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SEVERAL SPECIES OF DACTYLELLA AND DACTYLARIA THAT CAPTURE FREE-LIVING NEMATODES

CHARLES DRECHSLER [1]

(WITH 15 FIGURES)

Owing to their production of colorless pluriseptate conidia on tall colorless conidiophores, six clampless nematode-capturing hyphomycetes, evidently distinct from all previously described related forms of similar predacious character, are herein set forth as new species of *Dactylella* and *Dactylaria*. It is believed that the several species are distributed between the two genera in tolerable conformity with established usage, though as has been pointed out earlier (**13**: 467), the distinction between a solitary and a capitate sporulating habit is sometimes rather difficult to apply among members of the predacious series. The several new species came to light in agar plate cultures which after being overgrown with *Pythium* mycelium had been further planted with small quantities of decaying vegetable detritus from different localities; the decaying material in each instance supplying not only the nematodes that through rapid multiplication soon infested the agar abundantly but also the fungi by which the animals were subsequently destroyed in large numbers. Occasion is taken, besides, to set

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forth in a provisional way the predacious organs of a related nematode-capturing hyphomycete that so far has not been seen giving rise to reproductive apparatus. Some attention, furthermore, is given to a mucedinous form held referable to *Gonatobotrys simplex* Corda (**9**), since in its mycelial habit it offers suggestive resemblances to one of the six new species, and in its sporulating habit shows general parallelism more especially with the repeatedly nodose species of *Arthrobotrys* so widely active in the capture of eelworms.

A DACTYLELLA WITH SMALL CONSTRICTING RINGS AND MOSTLY BISEPTATE OR TRISEPTATE CONIDIA

Several Petri plates of maizemeal agar which after being overgrown with mycelium of *Pythium ultimum* Trow had been further planted with small quantities of leaf mold taken from the floor of pine (*Pinus* spp.) woods in Yellowstone National Park on August 18, 1947, became abundantly infested with eelworms during the ensuing weeks. By far the larger number of individual eelworms were conspicuously slender in shape and belonged, according to Dr. G. Steiner, to two species of *Plectus*. On microscopic examination 42 days after the leaf mold had been added the eelworms, especially those of the slender species, were found being killed over a wide area by a strangling hyphomycete differing in appearance from any known form of similar biological habit.

The vegetative mycelium of the hyphomycete came forth here and there from the deposits of opaque detritus to extend sparsely into the transparent agar. It was composed of sparingly branched colorless filaments partitioned at moderate intervals by cross-walls. At somewhat longer intervals the hyphae bore sturdy three-celled rings (FIG. 1, *A*; FIG. 2, *A*, *a*, *b*) which like the similar organs of other nematode-strangling forms were usually oriented in a plane perpendicular to the hyphal axis, and consequently were most often seen in edgewise view. The cellular make-up of the rings was better revealed in the occasional specimens that had been pushed sideways through the jostling of vigorous nematodes to be brought conveniently into lateral view (FIG. 1, *B–E*; *F*, *a*, *b*. FIG. 2, *B–E*); though such change in position entails some distortion of the stalk and at times also of the adjacent portion of mycelial filament. As

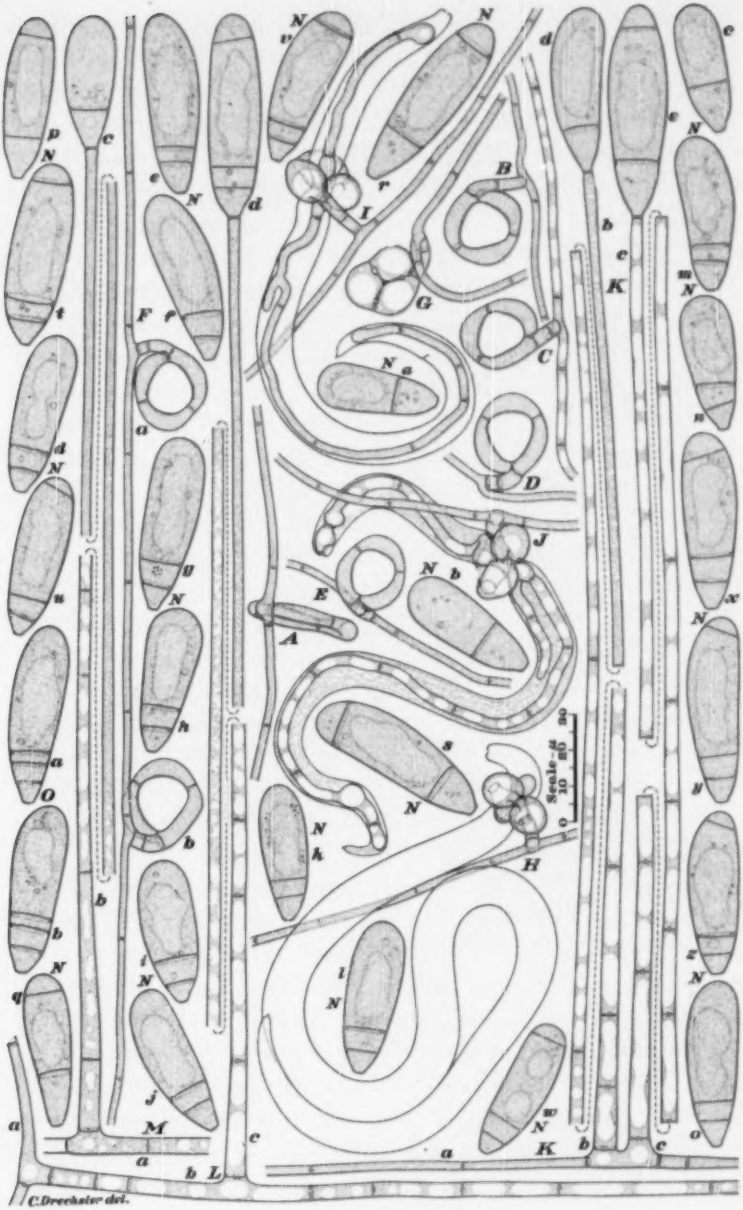

FIG. 1. *Dactylella stenobrocha.*

in other species the stalks were regularly composed of two cells. In relatively short stalks the proximal cell was often shorter than the distal cell (FIG. 1, *C; D; F, a, b*), thereby providing similarity to the strangling species I have described earlier under the binomials *Arthrobotrys dactyloides* (**12**: 482–487), *Dactylella bembicodes* (**12**: 487–492) and *Dactylaria brochopaga* (**12**: 514–518). Yet frequently, again, the two cells were of about equal length (FIG. 1, *B;* FIG. 2, *B–E*) somewhat as in my *Dactylella coelobrocha* (**19**). The rings themselves closely resembled the corresponding organs of *Arthrobotrys dactyloides, Dactylella bembicodes* and *Dactylaria brochopaga* in always showing smooth curvature along their rounded triangular inner profile as well as along their more nearly circular outer profile. Their three arcuate cells were never found showing at their inner profile the curious median bulge whereby the aperture of the constricting ring in my *Dactylella doedycoides* (**13**) and my *Dactylella heterospora* (**15**) is given a scalloped outline suggestive of trefoil ornamentation. They tapered moderately toward the cross-walls separating them, like the arcuate cells of most nematode-strangling species, and thus did not share the more pronounced taper characteristic of the arcuate cells in *Dactylella coelobrocha*. In general the rings here appeared of smaller and less variable size than those of any nematode-strangling form hitherto described with the exception possibly of my *Trichothecium polybrochum* (**12**: 535–538). The range of dimensions shown by them would in most allied species include only the constricting rings of small and medium sizes.

Here and there in the cultures where the fungus made its appearance rings were found that had closed emptily (FIG. 1, *G;* FIG. 2, *F*), thereby revealing advantageously the change of the three component cells from an elongated arcuate to an obese orbicular shape. Usually, however, the rings became closed in capturing individual specimens of the two slender species of *Plectus* (FIG. 1, *H–J.* FIG. 2, *G; H, a; I; J*) abundantly infesting the agar substratum. In some instances a ring that had closed emptily (FIG. 2, *H, b*) was found near one that held a captured nematode, suggesting that disturbance by the struggling captive, or perhaps impact of the animal's body against the flat side of the ring, may have supplied the stimulus that resulted in unprofitable closure. Captured

eelworms were always found indented about equally by all three swollen cells; the ring here manifestly operating in the manner most widely prevalent among nematode-strangling forms rather than in the manner distinctive of *Dactylella coelobrocha*, which commonly grips the animal between the first and second cells. As long as a captured eelworm continued struggling with some vigor, no change was observable near the constricted ring. Once the animal was capable only of feeble movements its integument was penetrated by one or more of the three swollen cells, and into its fleshy body globose protuberances were intruded forward and backward (FIG. 1, *H*). These protrusions soon grew out distally into assimilative hyphae which finally extended the invasion through virtually the whole length of the captive (FIG. 1, *I*). Frequently in very slender eelworms only a single assimilative hypha was extended forward and backward from the enveloping ring (FIG. 1, *I*), but in somewhat stouter animals the median portions usually came to be occupied by two hyphae (FIG. 1, *J*. FIG. 2, *H, a; J*). At the anterior end of the animal, as also at its tail end, the assimilative hyphae often showed marked distension in one or two terminal segments (FIG. 1, *I, J;* FIG. 2, *H, I*), a type of modification noted previously in *Arthrobotrys dactyloides* and *Dactylella coelobrocha*. During the progress of invasion and for some time afterwards the assimilative hyphae were obscured badly, owing to the globuliferous character of the animal's degenerating contents, but as these contents gradually diminished the assimilative hyphae emerged into view more and more clearly. When the animal's substance was in large part depleted the assimilative hyphae became noticeably vacuolate (FIG. 1, *J;* FIG. 2, *H, J*). The vacuoles increased steadily in volume on further reduction of the materials in the eelworm, until finally the hyphal envelopes were no less empty of living protoplasm than the animal's integument surrounding them.

With ample nourishment thus being obtained through expropriation of captured eelworms, the fungus produced conidiophores at variable intervals along the mycelial filaments (FIG. 1, *K, a; L, a*) on the surface of the agar substratum. The conidiophores (FIG. 1, *K, b, c*. FIG. 2, *K, a; L, a*) here consisted of sturdy erect septate hyphae, about 0.5 mm. high, that tapered very gradually toward the tip where each bore a solitary conidium (FIG. 1, *K, d, e*. FIG. 2,

K, b; L, b). After the conidium had been formed the conidiophore, while still in an erect posture, sometimes lost much (FIG. 3, *A*) or all (FIG. 3, *B*) of its protoplasmic contents. Sometimes, again, it would fall over on the substratum while all (FIG. 1, *L, b*) or some (FIG. 1, *M, a*) of its segments remained alive, and then in many instances would give rise, usually from one of its proximal segments, to a secondary conidiophore (FIG. 1, *L, c; M, b*) which, like its parent, bore a single conidium (FIG. 1, *L, d; M, c*).

The conidia produced by the fungus in nematode-infested agar cultures were regularly of elongated ellipsoidal shape. Many though not all of them were slightly wider in the distal portion than in the proximal portion. Usually the distal end was broadly rounded, whereas the lower half of the spore commonly tapered toward the somewhat rounded truncate base. They were divided variously by cross-walls ranging in number from one to three. In the uniseptate conidia (FIG. 1, *N, a–c;* FIG. 2, *M, a*), which were relatively infrequent, the septum nearly always delimited a long distal cell from a shorter proximal cell. The biseptate conidia, which were abundant, consisted of two short cells and one long cell; the long cell in some instances (FIG. 1, *N, d–o;* FIG. 2, *M, b–m*) surmounting the two short ones, and in other instances (FIG. 1, *N, p–s;* FIG. 2, *M, n, o*) being placed between them. The almost equally numerous triseptate conidia contained three short cells and one long cell; the long cell here most often occurring in penultimate position (FIG. 1, *N, t–z.* FIG. 2, *M, p–z; N*) between a short apical cell and two short proximal (*i.e.*, basal and parabasal) cells, though sometimes it occupied a terminal position (FIG. 1, *O, a, b*). In mature conidia, as a rule, the large cell contained a rather large elongated vacuole around which subspherical granules were often found scattered a little more thickly than elsewhere in the protoplasm. Conidia after falling on a moist substratum were often found sending up one (FIG. 2, *O, P*) or two (FIG. 2, *Q*) tapering aerial hyphae from any of the small cells. These aerial hyphae were noticeably more slender than ordinary germ hyphae extended by the conidia (FIG. 2, *R*) when immersed in fresh water or in some agar medium.

The fungus was isolated by removing conidia aseptically from the tall conidiophores to Petri plates of sterile maizemeal agar, the

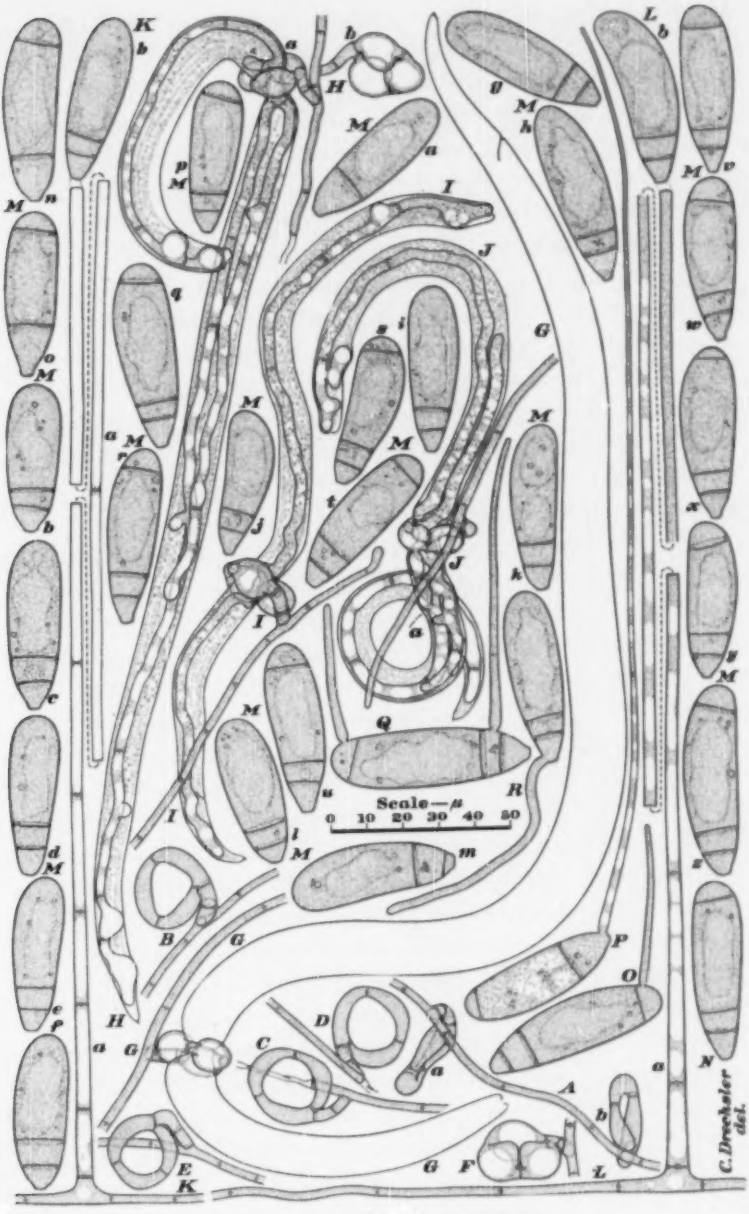

FIG. 2. *Dactylella stenobrocha.*

transfer being accomplished readily by touching the conidia with a small slab of sterile agar held on a flamed platinum spatula. In pure culture on maizemeal agar kept at a temperature of 20° C. it grows with moderate rapidity, usually without forming any predacious rings. Predacious rings corresponding accurately both in their cellular make-up and in their relatively small dimensions to those produced in nematode-infested materials have, however, been found present abundantly in pure cultures of the fungus in tubes of maizemeal agar that had been stored for over eleven months at a temperature of 5° C. in containers affording some protection against evaporation. As the aging cultures, on microscopical examination, showed no contamination by any bacteria, nor any admixture of other fungi, nor any infestation by animals, it would seem that the new hyphomycete can give rise to predacious rings, even though perhaps only rather slowly, without any chemical or physical stimulus from alien organisms.

Although the fungus sporulates rather freely in pure culture on maizemeal agar at temperatures near 20° C., the conidial apparatus produced here differs in dimensions from that formed on nematode-infested substratum. The conidiophores (FIG. 3, *C–E*) are usually much shorter, their height ranging ordinarily from 150 to 250 μ. The conidia likewise are shorter, commonly varying from 30 to 40 μ in length; and as their width is reduced only little, if at all, they are generally of smaller size and plumper shape. Uniseptate conidia (FIG. 3, *F*, *a–o*) are produced more frequently in pure culture, though showing usually the same manner of partitioning into a longer distal cell and a shorter proximal cell that is most prevalent in nematode-infested cultures. Biseptate conidia here, much as in nematode-infested cultures, may have both cross-walls placed toward the basal end (FIG. 3, *G*, *a–h*) or may show a more symmetrical arrangement in having one cross-wall near the basal end and the other near the distal end (FIG. 3, *G*, *i–p*). Triseptate conidia (FIG. 3, *H*), if formed less frequently than in nematode-infested cultures, usually display similar disposition of cross-walls. The fungus evidently sporulates also at relatively low temperatures, since in pure culture in tubes of maizemeal agar stored at 5° C., it showed on examination after eleven and one half months an abundance of new living conidia. These conidia seemed

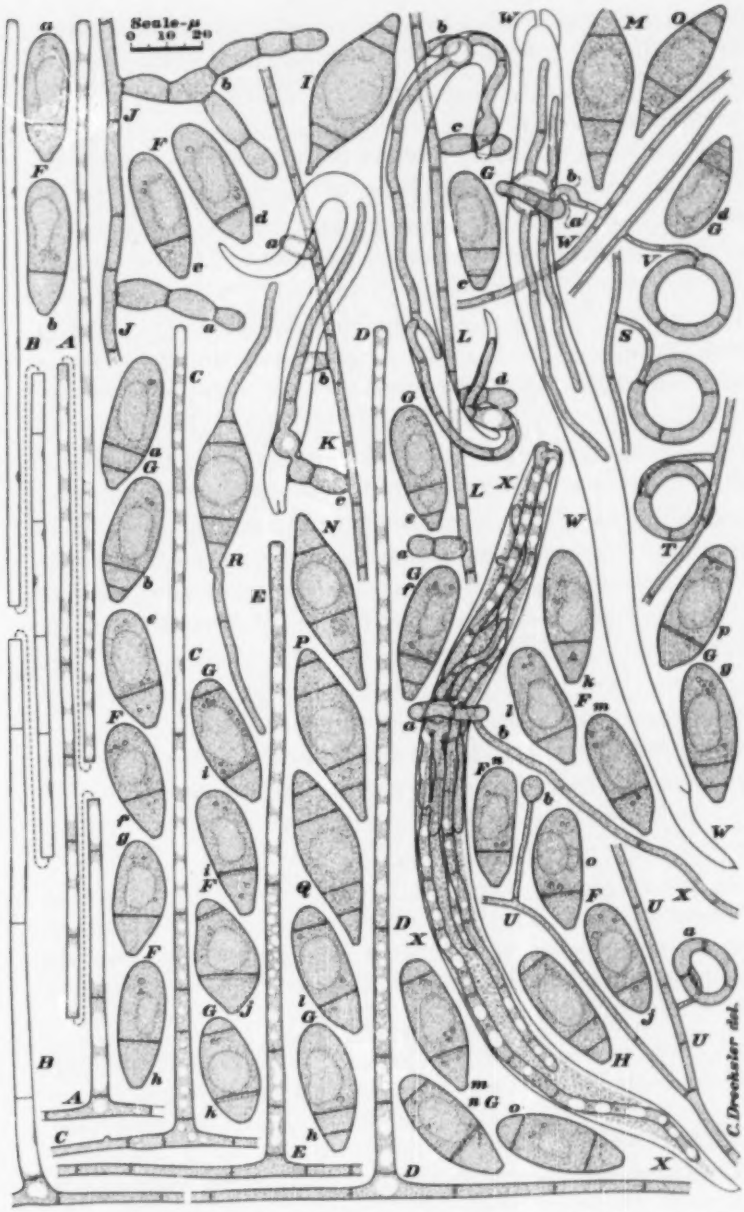

FIG. 3. *A–H, Dactylella stenobrocha; I, D. aphrobrocha; J–R, D. cionopaga; S–X,* Unnamed Hyphomycete.

generally larger than those produced at 20° C., and were given more frequently to rather pronounced curvature. The conidiophores had a tendency toward production of successive spores following repeated elongation.

Despite the promiscuous variations of its conidia in pure culture, the fungus was never found producing spores that could be regarded as belonging in a different category from the elongated ellipsoidal conidia produced in nematode-infested cultures. It was never observed giving rise to any supplementary reproductive bodies comparable, for example, to the uniseptate allantoid conidia of *Dactylella heterospora*, or to the elongated globuliferous spores of *Dactylella doedycoides*. It has never been seen provided with chlamydospores such as are present abundantly in cultures of *Arthrobotrys oligospora* Fres., nor with distinctive aggregations of enlarged indurated mycelial cells of the sort familiar in cultures of *Dactylella heterospora*, though after several months some of its wider hyphae often appear slightly indurated in that their walls are then thickened and their contents have become partly globulose. While its conidia, in their elongated shape, most nearly resemble the large regularly biseptate conidia of *Dactylella heterospora*, they have never been found produced in submerged positions under the surface of agar substrata, their contour at the base is rounded truncate rather than sharply truncate, and they frequently contain three cross-walls—so frequently, indeed, that the triseptate condition may well be regarded as being typical of the species. In their outward shape they differ markedly from the obese conidia of *Dactylella doedycoides*, as well as from the swollen turbinate conidia of *Dactylella bembicodes;* while their generally greater dimensions and different arrangement of septa distinguish them from the conidia of *Dactylaria brochopaga*, which are usually borne in capitate clusters rather than singly.

A term having reference to its relatively small constricting rings is deemed suitable as a specific epithet for the fungus.

Dactylella stenobrocha sp. nov.

Mycelium sparsum; hyphis hyalinis, mediocriter septatis, plerumque 1.7–3.7 μ crassis, hic illic ex ramulis bilocularibus (rarius trilocularibus) vulgo 8–14 μ longis et 3–4.5 μ crassis laqueos circulares 20–31 μ (vulgo circa 23 μ) latos proferentibus qui in 3 cellulis arcuatis 14–25 μ (vulgo circa 17 μ) longis

medio 4.4–5.5 μ extremo circa 3 μ crassis consistunt et foramen rotundum vel rotundo-triangulum 11–21.5 μ (vulgo circa 13 μ) latum circumdant; vermiculo nematoideo in laqueum apertum errato omnibus tribus cellulis abrupte se contrahentibus, itaque animal captivum magnopere comprimentibus, mox id trucidentibus, integumentum ejus perforantibus, hyphas intus evolventibus quae carnem exhauriunt; hyphis assumentibus incoloratis, septatis, magnam partem 2–3.5 μ crassis, sed saepe in cellulas 4–8 μ crassas abeuntibus. Hyphae fertiles incoloratae, erectae, vulgo 425–550 μ altae, basi 4.5–6.5 μ crassae, sursum leniter usque 2.5–3 μ attenuatae, 3–10 septatae, apice quandoque leviter latescentes, ibi saepe 3–4.5 μ crassae, unum conidium ferentes; conidiis hyalinis, elongato-ellipsoideis vel late digitiformibus vel interdum aliquantulum clavatis, rectis vel leniter curvatis, basi aliquid rotunde truncatis, apice late rotundatis, vulgo 34–56.5 μ longis, 12.5–16.5 μ crassis, 1–3 septatis, in una grandiore cellula et 1–3 minoribus cellulis consistentibus, grandiore cellula (in uniseptatis sporis plerumque cellula superiore, in pluriseptatis sporis plerumque cellula ultima aut paenultima) vulgo 21–38 μ longa, minoribus cellulis vulgo 3–15 μ longis.

Vermiculos nematoideos diversos praecipue nematoidea gracilia capiens consumensque habitat in humo silvestri in Yellowstone National Park, Wyoming.

Mycelium scanty; vegetative hyphae colorless, septate at moderate intervals, mostly 1.7 to 3.7 μ wide, often especially in presence of nematodes producing mostly underneath and in perpendicular positions approximately circular rings measuring 20 to 31 μ (usually about 23 μ) in outside diameter and composed individually of three arcuate cells 14 to 25 μ (usually about 17 μ) long, 4.4 to 5.5 μ wide in the middle and about 3 μ wide at the ends—the first and third of the cells being united to each other as well as to the distal end of a frequently somewhat curved bicellular (occasionally tricellular) supporting stalk commonly 8 to 14 μ long and 3 to 4.5 μ wide, and all three of the cells having a smooth profile without median protuberance on the side bordering the ring aperture; this aperture being of circular or rounded triangular shape and varying commonly from 11 to 21.5 μ in diameter, most often being about 13 μ wide. After entrance of a nematode into the ring aperture all three arcuate cells abruptly contracting, thereby constricting the animal to death or disabling it, then perforating the integument and extending lengthwise through the body assimilative hyphae for the most part 2 to 3.5 μ wide but often terminating in cells 4 to 8 μ wide. Conidiophores colorless, erect, commonly 425 to 550 μ high, 4.5 to 6.5 μ wide at the base, tapering upward very gradually to a distal width of 2.5 to 3 μ, ultimately becoming divided by 3 to 10 cross-walls, at the apex sometimes widening slightly to a diameter of 3 to 4.5 μ, and bearing there a single conidium. Conidia colorless, elongate elliptical or broadly finger-shaped or sometimes slightly clavate, straight or slightly curved, somewhat rounded truncate at the narrowed base, broadly rounded at the tip, commonly

34 to 56.5 μ long and 12.5 to 16.5 μ wide, divided by 1 to 3 cross-walls into 2 to 4 cells whereof one exceeds the other or others in size, the larger cell (in uniseptate spores usually the distal cell, in pluriseptate spores usually either the apical or the penultimate cell) commonly 21 to 38 μ long, the smaller cells commonly 3 to 15 μ long.

Capturing and consuming different nematodes, especially nematodes of slender body shape, it occurs in leaf mold in Yellowstone National Park, Wyoming.

A DACTYLELLA PRODUCING LONG-STALKED CONSTRICTING RINGS AND QUADRISEPTATE CONIDIA

Six years ago a nematode-strangling fungus resembling *Dactylella bembicodes* in the general appearance of its tall conidiophores and solitary conidia was found developing meagerly in a maizemeal-agar plate culture which after being overgrown with mycelium of my *Pythium salpingophorum* had been further planted with a small quantity of leaf mold gathered in a deciduous wood near Fairfax, Virginia, on November 10, 1942. The fungus arrested attention because its conidia, though of broadly fusiform shape much like those of *D. bembicodes*, were nearly always divided by four rather than by three cross-walls (FIG. 3, *I*). As *Dactylella coelobrocha* had not then been discovered the quadriseptate condition was quite alien to the conidia of any nematode-strangling hyphomycete known at the time. It was noted, moreover, that the stalks bearing the constricting rings were in general a little longer than in *D. bembicodes*. To determine whether the observed differences came about as variations of *D. bembicodes* or derived from a separate species, attempts were made to isolate the fungus by removing its conidia to a sterile agar medium. These attempts failed owing to the unfortunate circumstance that the rather small number of conidiophores available were closely intermingled with taller sporangiophores of a species of *Mucor*. Every lot of conidia removed was in some measure mixed with small spores of the *Mucor*. In all cultures planted with the mixture, the rapidly growing *Mucor* quickly overwhelmed the hyphomycete.

Early in 1948 the same fungus developed abundantly in more than a dozen maizemeal-agar plate cultures which, after being perme-

ated with mycelium of *Pythium debaryanum* Hesse, had been further planted with small quantities of leaf mold kindly gathered by Dr. E. B. Toole in woods consisting mainly of pine (*Pinus*) and oak (*Quercus*) trees near Greensboro, North Carolina, on December 29, 1947. From the lofty conidiophores sent up in scattered positions over extensive tracts of substratum in these cultures, conidia of the fungus were readily transferred to sterile agar without admixture of any alien organism. Ample opportunity was thus provided for comparing the fungus, in pure as well as in nematode-infested cultures, with all known hyphomycetes of similar predacious habit, apart from *Trichothecium polybrochum*, which has not been seen again since it was first encountered in 1933. The fungus from North Carolina was soon revealed as differing decisively from *Dactylella bembicodes* and *Dactylella coelobrocha*. Through its disclosure as a separate species the number of distinct mucedinous forms known to utilize constricting rings in their capture of eelworms is increased to nine.

When sizable portions are cut from pure cultures of the new *Dactylella* and placed on nematode-infested agar in Petri dishes, mycelial filaments grow out somewhat sparingly in all directions from the transferred material into the surrounding medium. While the mycelium developed by the fungus in pure culture on maizemeal agar is regularly free of predacious organs, the hyphae extended into nematode-infested agar promptly give rise to constricting rings at somewhat variable intervals. These rings, much as in allied species, are commonly formed in planes perpendicular or nearly perpendicular to the mycelial filament bearing them (FIG. 4, *A*; *B*, *a*. FIG. 5, *A*). Some of them, on being jostled vigorously by robust eelworms, are turned sideways in noticeable measure (FIG. 4, *B*, *b*; *C*, *a*, *b*; *D*; *E*. FIG. 5, *B*), and a few are turned flatwise (FIG. 4, *F–H*; *I*, *a*, *b*. FIG. 5, *C–H*) so that their cellular make-up is conveniently exposed to view. On several occasions it was possible to observe progressive steps in the development of predacious rings oriented flatwise in slabs of nematode-infested agar mounted on a microscope slide under a cover glass. The developing organ in each instance first became recognizable as a circinately curved lateral branch delimited by a basal septum from the frequently somewhat narrower parent hypha (FIG. 5, *I*, *a*), and filled with

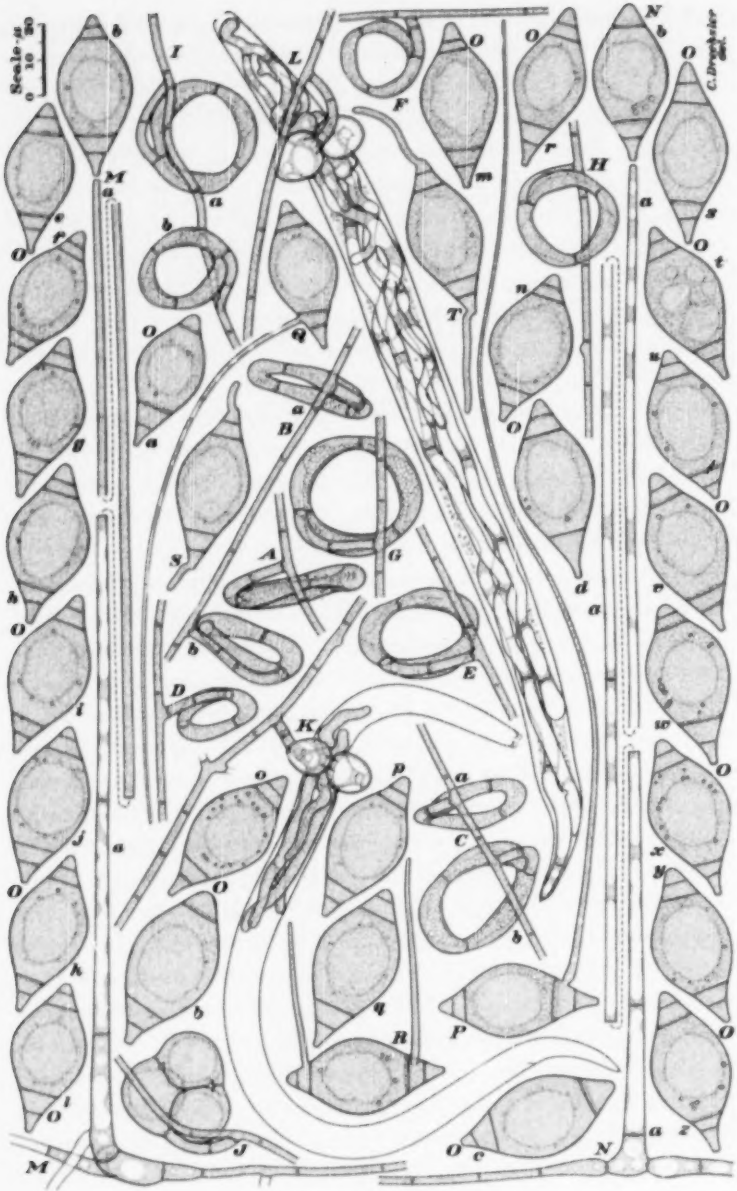

FIG. 4. *Dactylella aphrobrocha.*

protoplasm of nearly homogeneous consistency. Two cross-walls soon appeared in the proximal portion of the branch to delimit the two segments destined to make up the stalk. As the tip of the recurving branch approached the farther septum a spur grew out from the distal end of the second stalk cell to meet it (FIG. 5, *I*, *b*). The spur often attained a length of 3 or 4 μ before its tip encountered the tip of the branch (FIG. 5, *I*, *c*). At about the same time another cross-wall was formed to delimit the proximal arcuate segment of the ring that came into being as the juxtaposed tips fused broadly (FIG. 5, *I*, *d*). Later a second connection was established through hyphal fusion between the basal end of the proximal arcuate segment and the portion of the ring that had originated as a lateral spur. The continuity of both anastomosing connections eventually was modified through the formation of a cross-wall in each; one of the cross-walls being laid down in a position corresponding to the base of the lateral spur, whereas the other was laid down midway in the second connection. With the deposition meanwhile of another cross-wall midway between the distal end of the first arcuate segment and the septum formed at what had been the base of the lateral spur, the second and third arcuate segments were delimited from one another, thereby completing the cell divisions required in putting forth a constricting ring of usual structure (FIG. 5, *I*, *e*).

Departure from the usual cellular structure is observable now and then in instances where the stalk consists of three (FIG. 4, *B*, *b*; FIG. 5, *J*) rather than two cells. These three-celled stalks commonly exceed in length the arcuate segments of the rings supported by them, whereas the bicellular stalks are most often a little shorter than the associated arcuate segments. However, even the bicellular stalks must be considered relatively long in comparison with those of most allied species, being about equal in length to the rangy stalks of *Dactylella doedycoides* and *Dactylella heterospora*. The arcuate cells in the present fungus lack the median bulge whereby the aperture of the rings in *Dactylella doedycoides* and *Dactylella heterospora* is given a scalloped outline suggestive of trefoil ornamentation. They taper toward their delimiting septa in about the same measure as the homologous cells of all other known nematode-strangling forms apart from *Dactylella coelobrocha*. Apparently the arcuate cells acquire their definitive shape together with their

contractile power during their later stages of development. During these later stages (FIG. 5, *I, d, e*) they increase perceptibly in length as the ring enlarges, while their thickness near the septa increases in about the same proportion. In their median region, however, they undergo more pronounced widening, so that their width here often comes to be twice that of the stalk cells with which they had formed earlier a branch of fairly uniform diameter. Distinctive changes take place also in their internal structure. Arcuate cells capable of contraction show along the inner side bordering the ring aperture a rather thin layer of protoplasmic material having a characteristic dense appearance. Material of similarly dense consistency is likewise present in a layer of more variable thickness along the outer side. Between the two dense layers is an irregular elongated region filled with rather faintly visible globules which collectively offer a somewhat foamy appearance when the ring is viewed flatwise. When the ring is viewed edgewise in its normal perpendicular posture the arcuate cells appear as if they were marked by a series of curved transverse striations (FIG. 4, *A*). Presumably the globuliferous core here corresponds to the elongated vacuole or vacuole-like part displayed by the arcuate cells of *Dactylella coelobrocha*.

In nematode-infested cultures predacious organs may be found here and there that have closed emptily (FIG. 4, *J*), thus showing conveniently the pronounced contraction undergone by the three component cells in changing from an arcuate to an orbicular shape. Ordinarily, of course, closure of the organ results in the capture of a nematode; the animal being gripped most often at its anterior end (FIG. 4, *K; L;* FIG. 5, *K–M*), though occasionally it is held by the tail end (FIG. 5, *N*). All three swollen cells of the closed ring are indented broadly and deeply into the yielding body of the eelworm. After the captive's struggles have become feebler, presumably from exhaustion, its integument is narrowly perforated in two or three places by slender protuberances from the swollen cells. Each protuberance, on reaching the fleshy interior, widens markedly (FIG. 5, *K*) to become recognizable as an assimilative hypha (FIG. 4, *K*). The several assimilative hyphae elongate until the animal's body is invaded from head to tail (FIG. 4, *L;* FIG. 5, *L, N*). During their period of active growth the assimilative hyphae show only

rather scanty septation, but since they continue to form cross-walls for some time after growth ceases, they eventually are divided into segments mostly 10 to 50 μ long. In some instances the main assimilative filaments bear a moderate number of branches and spurs (FIG. 5, *M*), but in other instances, again, branching is infrequent (FIG. 4, *L*; FIG. 5, *N*), or even wholly absent (FIG. 5, *L*). Hyphal fusions between assimilative hyphal elements are to be found occasionally (FIG. 5, *M*). Although now and then an assimilative hypha terminates in a noticeably expanded cell (FIG. 4, *L*), this sort of modification would seem hardly frequent enough to be regarded as a characteristic feature of the species.

The assimilative hyphae in their progressive invasion of a captured eelworm bring about globulose degeneration of its fleshy substance, and thereby are badly obscured from view, especially if the animal's body is relatively stout. After the globulose materials have been largely absorbed by them, the hyphae become more clearly visible. On further depletion of the animal's substance, vacuoles make their appearance in the hyphal segments (FIG. 4, *L*; FIG. 5, *N*). With continued enlargement of these vacuoles, the protoplasmic contents of the assimilative hyphae are steadily withdrawn backward into the mycelial filament bearing the predacious organ, until ultimately the hyphal envelopes, like the collapsing integument surrounding them, are left entirely empty. The transfer of hyphal contents backward into the mycelial filament is sometimes accomplished by way of the stalk on which the constricting ring was produced (FIG. 4, *L*). Often, however, the original stalk degenerates (FIG. 5, *M*, *a*; *N*, *a*), very probably as the result of injury sustained from the violent struggles of the captured eelworm. Where such degeneration occurs a new hyphal element is produced to provide a passageway for the backward movement of fungus protoplasm. Usually the new hyphal element (FIG. 5, *M*, *b*) connects the mycelial filament with one or another of the three swollen cells. Somewhat rarely, where the swollen cells have all suffered some injury, the new connection may be established directly between the mycelial filament and the proximal hyphal cell within the animal (FIG. 5, *N*, *b*). In its ready development of new hyphal connections the fungus shows obvious similarity to *Dactylella doedycoides* and *Dactylella heterospora*, which, as has been noted, likewise bear

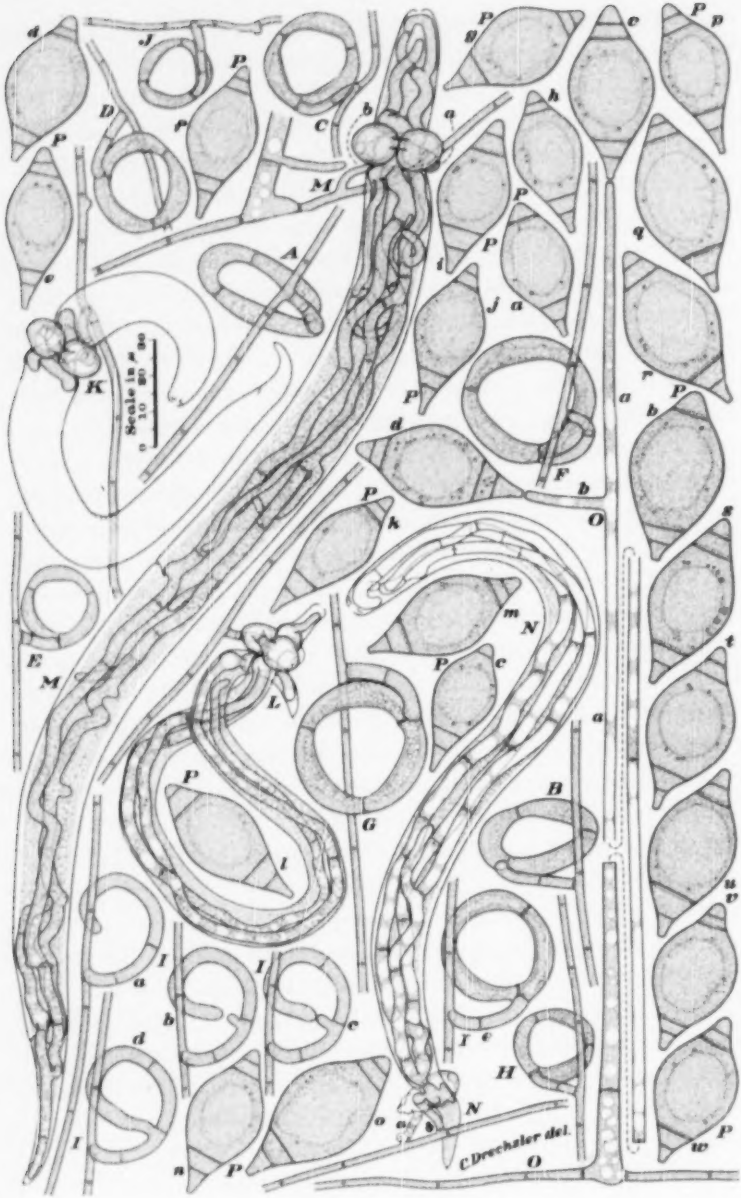

FIG. 5. *Dactylella aphrobrocha.*

their predacious rings on relatively long stalks easily injured by violent struggles of captured eelworms.

The scattered erect conidiophores sent up by the fungus from hyphae prostrate on nematode-infested substrata resemble those of *Dactylella bembicodes* in their stature and gradual upward tapering (FIG. 4, *M, a; N, a*). As a rule they give rise terminally to a single conidium (FIG. 4, *M, b; N, b*), but now and then a conidiophore (FIG. 5, *O, a*) will put forth a lateral branch (FIG. 5, *O, b*) some distance below the tip, and thus is enabled to form two conidia (FIG. 5, *O, c, d*). Although on the whole the conidia here are somewhat longer and wider than those of *Dactylella bembicodes*, and in general appear to have their greatest width rather more accurately midway between base and apex, the differences in size and shape they present in comparison with the widely distributed constricting species made known earlier are not very pronounced. In some instances they contain three cross-walls, which often delimit a small apical cell, a large ventricose penultimate cell, a small parabasal cell, and a small basal cell (FIG. 4, *O, a–c;* FIG. 5, *P, a–c*), thus bringing about the usual cellular arrangement found in spores of *Dactylella bembicodes*. Often again, however, in triseptate conidia it is the parabasal cell that is large and ventricose, while the penultimate cell, as also the apical and basal cells, will be relatively small (FIG. 4, *O, d*). Although triseptate conidia are formed rather abundantly in pure culture on maizemeal agar, by far the greater number of conidia produced by the fungus in nematode-infested cultures are symmetrically quadriseptate (FIG. 4, *O, e–z;* FIG. 5, *P, d–w*); a massive ventricose cell with a large vacuole being found in median position between two smaller proximal (basal and parabasal) cells below and two smaller distal (penultimate and apical) cells above. As conidia with more than four cross-walls are virtually unknown here, the quadriseptate condition may safely be regarded as being definitive in mature spores, whereas the positions of the cross-walls in many triseptate specimens taken from actively sporulating nematode-infested cultures give ample reason for suspecting that had no disturbance intervened a fourth transverse partition would have been formed. Accordingly the data on conidial dimensions given in the diagnosis were derived, much as in the case of *Dactylella coelobrocha*, mainly from quadri-

septate specimens taken at random in mounts prepared from abundantly sporulating nematode-infested agar plate cultures. One hundred measurements for length, expressed in the nearest integral number of microns, showed a distribution as follows: 41 μ, 1; 42 μ, 2; 43 μ, 1; 44 μ, 15; 45 μ, 13; 46 μ, 21; 47 μ, 17; 48 μ, 11; 49 μ, 6; 50 μ, 3; 51 μ, 6; 52 μ, 2; 54 μ, 1; 55 μ, 1. The measurements for greatest width, relating to the same 100 conidia, were distributed thus: 17 μ, 1; 18 μ, 1; 19 μ, 7; 20 μ, 7; 21 μ, 21; 22 μ, 22; 23 μ, 25; 24 μ, 12; 25 μ, 3; 26 μ, 1; while the median cells of the 100 specimens gave values for length as follows: 23 μ, 2; 24 μ, 1; 25 μ, 7; 26 μ, 20; 27 μ, 25; 28 μ, 23; 29 μ, 14; 30 μ, 6; 31 μ, 1; 34 μ, 1. Although the conidia here are substantially shorter than those of *Dactylella coelobrocha*, the two species show rather little difference with respect to the lengths of the parabasal, median, and penultimate cells. Manifestly the conidia of *Dactylella coelobrocha* owe their greater length, as also their markedly different appearance, to their characteristically protracted basal and apical cells, which are conspicuously longer than the concomitant parabasal and penultimate cells, respectively. Often the conidia of the present fungus, while still borne aloft on the conidiophores, will put forth a slender aerial hypha, usually from the small parabasal cell (FIG. 4, *P*, *Q*), but sometimes also from the small penultimate cell (FIG. 4, *R*). In general these aerial hyphae resemble the slender outgrowths extended by the conidia of *Dactylella coelobrocha*, but are perhaps somewhat less rigid. As in related species they seem to be produced more profusely under warm than under cool conditions. When mature conidia fall on a moist substratum or are shallowly immersed in water, they germinate in commonplace manner by putting forth a germ hypha from the small apical cell and from the small basal cell (FIG. 4, *S*, *T*).

In recalling the globuliferous core of its contractile arcuate cells a term compounded of two words meaning "foam" and "noose," respectively, may serve as a suitable specific epithet for the fungus.

Dactylella aphrobrocha sp. nov.

Mycelium sparsum; hyphis hyalinis, septatis, plerumque 1.7–3.7 μ crassis, hic illic ex ramulis bilocularibus (raro trilocularibus) vulgo 11–28 μ longis et 2.4–4.7 μ crassis laqueos circulares 20–38 μ latos proferentibus qui in 3

cellulis arcuatis consistunt et foramen rotundum vel rotundo-triangulum 12–27 μ latum circumdant; cellulis arcuatis 15–37 μ longis, medio 4–8.6 μ crassis, extremo 2.4–4.7 μ crassis, denso eorum protoplasmate circum lacunam centralem elongatam globuliferam disposito; vermiculo nematoideo in laqueum apertum introito omnibus tribus cellulis abrupte se contrahentibus, animal magnopere comprimentibus, id ita necantibus, integumentum ejus perforantibus, hyphas intus evolventibus quae carnem exhauriunt; hyphis assumentibus mox mediocriter septatis, saepius plus minusve ramosis, maximam partem 2–5 μ crassis sed interdum in cellulis usque 7 μ crassis abeuntibus, quandoque inter se conjunctis. Hyphae fertiles incoloratae, erectae, saepe 450–525 μ altae, basi 6–7 μ crassae, sursum leniter attenuatae, apice circa 2 μ crassae, ibi unum conidium ferentes, quandoque ramo brevi subter apicem praeditae denique aliud conidium gignentes; conidiis incoloratis, plerumque late fusiformibus, 2–4 septatis, saepissime quadriseptatis, tum plerumque ex toto 41–55 μ (saepius circa 46.7 μ) longis et 17–26 μ (saepius circa 21.9 μ) crassis, cellula infima 4.2–9.1 μ (saepius circa 6.2 μ) longa, cellula secunda 3.2–7.6 μ (saepius circa 5 μ) longa, cellula media 22.8–34 μ (saepius circa 27.3 μ) longa, cellula paenultima 2.6–6.5 μ (saepius circa 4.1 μ) longa, cellula summa 1.8–6 μ (saepius circa 4.1 μ) longa; cellula secunda interdum etiam rarius cellula paenultima appendicem filiformem ad pares angulos emittente, appendice incolorata, recta vel curvata, continua vel 1–2 septata, 40–300 μ longa, basi 1.8–2.5 μ crassa, sursum attenuata, apice circa 0.8 μ crassa.

Vermiculos nematoideos varios capiens consumensque habitat in humo silvestri prope Greensboro, North Carolina.

Mycelium scanty, spreading; vegetative hyphae colorless, septate, mostly 1.7 to 3.7 μ wide, often, especially in presence of nematodes, giving rise on curving stalks to circular rings in usually perpendicular positions; the stalks, commonly 11 to 28 μ long and 2.4 to 4.7 μ wide, consisting usually of two cells but occasionally composed of three cells; the rings, commonly 20 to 38 μ in outside diameter and surrounding a circular or rounded triangular aperture 12 to 27 μ wide, being consistently composed of three arcuate segments; the arcuate cells usually 15 to 37 μ long, 4 to 8.6 μ wide in the middle and 2.4 to 4.7 μ wide at the ends, containing dense homogeneous protoplasm in a parietal layer surrounding a central elongated lacuna filled with numerous globules—the first and third of the cells being united to one another as well as to the distal end of the stalk, and all the cells lacking a median protrusion on the inner side; on entrance of a nematode into the aperture the arcuate cells contracting abruptly, all three indenting the animal broadly and deeply, thereby strongly constricting it and soon disabling it, then perforating its integument to extend lengthwise through its body assimilative hyphae that appropriate the fleshy contents; the assimilative hyphae for the most part 2 to 5 μ wide but sometimes terminating in cells up to 7 μ in width. Conidiophores colorless, erect, often 450 to 525 μ high, 6 to 7 μ wide at the base, tapering

gradually upward, about 2 μ wide at the tip, there bearing a single conidium, and sometimes bearing another conidium on a short branch given off a little below the tip. Conidia colorless, usually broadly spindle-shaped, sometimes 2- or 3-septate, but most often divided by four cross-walls in such wise that the middle cell is much larger than the others—then mostly 41 to 55 μ (average 46.7 μ) long and 17 to 26 μ (average 21.9 μ) wide, with the basal cell measuring often 4.2 to 9.1 μ (average 6.2 μ) in length, the parabasal cell 3.2 to 7.6 μ (average 5 μ), the median cell 22.8 to 34 μ (average 27.3 μ), the penultimate cell 2.6 to 6.5 μ (average 4.1 μ), and the apical cell 1.8 to 6 μ (average 4.1 μ); the small parabasal cell, and occasionally also the small penultimate cell, under some conditions putting forth at a right angle with the spore axis an aerial hyphal appendage; the appendage being colorless, straight or flexuous, 40 to 300 μ long, 1.8 to 2.5 μ wide at the base, tapering gradually to a width of 0.8 μ at the tip, continuous or divided in the broader proximal portion by one or two cross-walls.

Capturing eelworms of different species it occurs in leaf mold in woods near Greensboro, North Carolina.

A DACTYLELLA WITH ADHESIVE COLUMNAR OUTGROWTHS THAT SOMETIMES UNITE TO FORM IRREGULAR MESHES

In some maizemeal-agar plate cultures which after having been overgrown with *Pythium* mycelium had been further planted with leaf mold collected in deciduous woods near Butternut, Wisconsin, in September 1938, there appeared among various other nematode-capturing hyphomycetes a *Dactylella* that in a general way resembled *D. gephyropaga*, yet with respect to several phases of morphology differed rather markedly from that species. Its mycelial hyphae bore columnar outgrowths that not infrequently were composed of three or more cells (FIG. 3, *J, a, b*), though most often they consisted of one or two cells (FIG. 3, *K, a, b; L, a*). These outgrowths, like those of *D. gephyropaga*, were operative in capturing eelworms through adhesion (FIG. 3, *K, c; L, b–d*), the captives soon being disabled from intrusion of globose infective bodies and then undergoing invasion by assimilative hyphae. The fungus produced conidia which usually contained three (FIG. 3, *M–O*) or four (FIG. 3, *P, Q*) cross-walls, but were noticeably longer than the triseptate and quadriseptate conidia characteristic of *D. gephyropaga*. After falling on a moist substratum the conidia often ger-

minated in a commonplace manner by putting forth a germ hypha from each end (FIG. 3, *R*).

The same fungus came to light again a few years later in maizemeal-agar plate cultures which after being overgrown with *Pythium irregulare* Buism. had been further planted through addition of small quantities of leaf mold gathered on October 2, 1941, near Presque Isle, Maine, in woods consisting largely of poplar (*Populus* sp.) and beech (*Fagus grandifolia* Ehrb.) trees. More recently it reappeared in more than a dozen maizemeal-agar plate cultures which after being permeated with *Pythium* mycelium had been planted with small quantities of wheat (*Triticum aestivum* L.) straw kindly collected by Dr. W. J. Zaumeyer near Hermiston, Oregon, on August 20, 1947. In these later cultures the distinctive characteristics of the fungus were brought more clearly into relief through the circumstance that nearly at the same time wholly typical material of *Dactylella gephyropaga* was found developing abundantly in nematode-infested agar plate cultures planted with leaf mold collected near Greensboro, North Carolina, and near Farmer, North Carolina, late in December, 1947. On transferring conidia aseptically from erect sporophores to tubes of sterile maizemeal agar the Oregon hyphomycete was obtained in pure culture. Comparison with pure cultures of *D. gephyropaga* originating from the two aforementioned localities in North Carolina as well as from several localities in Maryland and Virginia revealed it as a separate species of the same genus. Since the new *Dactylella* here in question has so far been obtained only from northern localities, and has never been recognized in the numerous cultures which in the course of 15 years were prepared with decaying detritus from many different places near Washington, D. C., there is reason to suspect that it may be distributed in colder regions than *D. gephyropaga*.

In the laboratory, certainly, the new fungus shows adaptation to lower temperatures than *Dactylella gephyropaga*. When portions of agar newly permeated with its mycelium are excised from pure cultures and placed on nematode-infested substratum kept at temperatures between 25° and 30° C., scarcely any hyphae are extended from the transferred mycelium, and virtually no eelworms will be captured, though within this temperature range under like conditions *D. gephyropaga* promptly sends out mycelial hyphae in

all directions, and on them produces columnar outgrowths and scalariform networks that operate with spectacular efficiency in the capture of nematodes. When similar preparations are kept at temperatures between 15° and 18° C., however, the two fungi show reversed capacity for predacious activity; the new species soon extending many mycelial filaments well beset with columnar outgrowths effective in capturing eelworms, whereas *D. gephyropaga* will remain almost wholly inactive. Owing to its different temperature adaptations and its departures in morphology the new *Dactylella* compares with *D. gephyropaga* in much the same way as my *Dactylaria psychrophila* (**16**: 154–163) compares with the apparently more widespread retiary hyphomycete I described earlier under the binomial *Dactylaria thaumasia* (**12**: 518–523).

Unlike *Dactylella gephyropaga*, the Oregon strain of the new *Dactylella* (FIG. 6; FIG. 7), as also the strains from northern Wisconsin and northern Maine, often gives rise to predacious organs about as freely when growing undisturbed in pure culture on maize-meal agar (FIG. 6, *A–F*; FIG. 7, *A–C*) as when developing on nematode-infested substratum. In either type of culture some of the predacious organs consist individually of a short, stout, dome-shaped or columnar unicellular outgrowth filled uniformly with dense protoplasm of finely granular texture (FIG. 6, *A*; *B*; *C*, *a*; *E*, *a*. FIG. 7, *B*, *a*). The columnar outgrowths of somewhat greater length are commonly pluricellular, consisting of two (FIG. 6, *C*, *b*, *c*; *E*, *b*. FIG. 7, *A*, *a*; *B*, *b*; *D*, *a*), three (FIG. 7, *A*, *b*), four (FIG. 6, *D*, *a*; *E*, *c–e*), or more (FIG. 6, *F*) cells. Here and there two bicellular outgrowths may be found united distally by a unicellular bridging segment (FIG. 6, *C*, *d*) to form a rectangular mesh similar in shape and cellular make-up to the meshes of *Dactylella gephyropaga*. In addition, meshes of rectangular shape occur that show only little departure from the design characteristic of *D. gephyropaga* in having the columnar outgrowths united distally by a two-celled (FIG. 7, *B*, *c*) rather than by a one-celled bridging connection. On the whole, however, union between columnar outgrowths is much less frequent than in *D. gephyropaga*, and takes place in a more haphazard manner so that for the most part meshes are formed singly in promiscuously scattered positions, with comparatively little uniformity of size, shape, or cellular composition (FIG. 6, *D*, *b*,

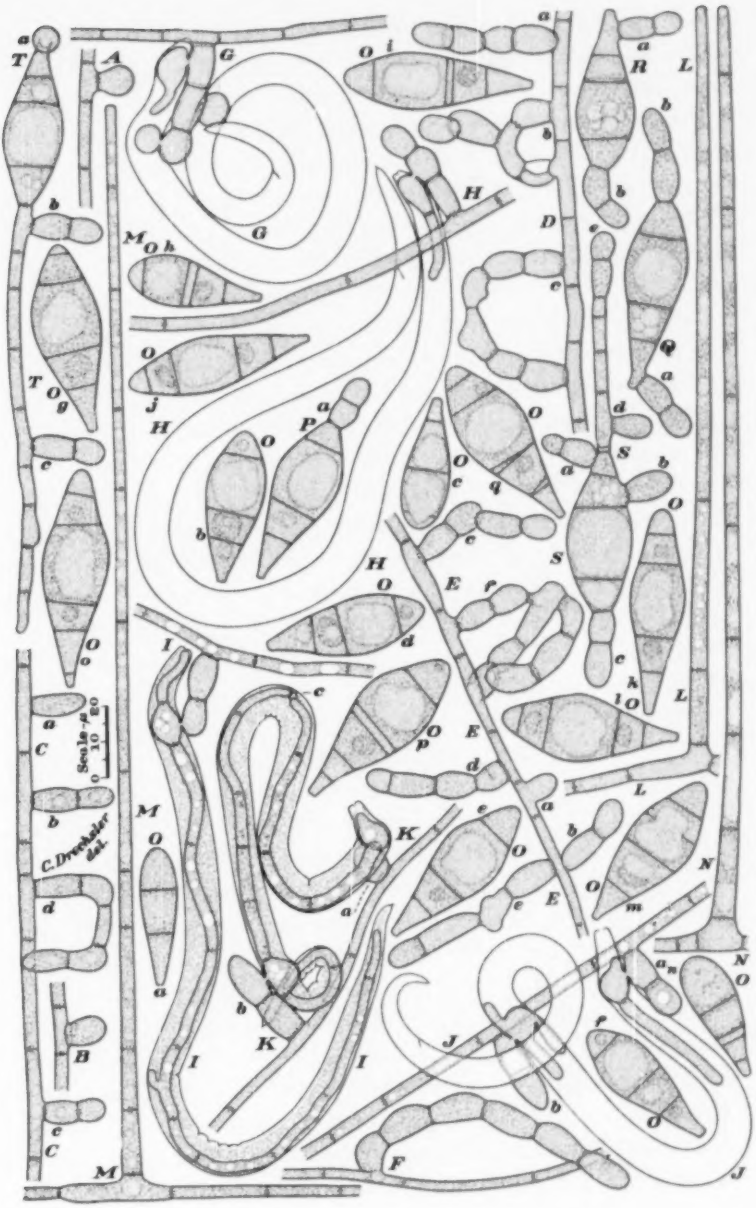

FIG. 6. *Dactylella cionopaga.*

c; *E*, *f*. FIG. 7, *C*). The arrangement of numerous meshes into extended scalariform networks, so frequent in *D. gephyropaga*, is not characteristic of the present fungus.

Since the new species forms closed meshes in lesser number than *Dactylella gephyropaga*, its capture of nematodes is accomplished almost wholly through adhesion. Very frequently only a single outgrowth is operative in holding an eelworm (FIG. 6, *G–I*; FIG. 7, *E–H*), though often, again, two or more outgrowths (FIG. 6, *J*, *a*, *b*; *K*, *a*, *b*. FIG. 7, *D*, *b*, *c*) are found participating. No coating of adhesive material is to be seen on the undisturbed outgrowth, but after a struggling animal has been held fast for some time a sizable cushion of colorless glutinous substance often becomes visible, especially in instances where the contact between fungus and eelworm is conveniently presented in profile view (FIG. 6, *K*, *b*; FIG. 7, *E*). Soon after capture is effected the animal's integument is narrowly perforated by one or more slender protrusions put forth by the fungus. Where pluricellular outgrowths are operative a protrusion may be extended from any or all of the component segments. Thus in the case of bicellular outgrowths, sometimes the distal segment (FIG. 6, *I*; *K*, *b*. FIG. 7, *D*, *c*; *E*) and sometimes the proximal segment (FIG. 6, *J*, *a*, *b*; FIG. 7, *F*, *G*) is directly active in penetrating into the captive; while with three-celled outgrowths penetration may take place from the median segment (FIG. 6, *H*), from the basal and apical segments (FIG. 7, *H*), or from all segments (FIG. 6, *G*). Once the integument is penetrated the narrow protrusion gives rise within the fleshy interior to a globose infective body (FIG. 6, *G*) that continues to enlarge until it occupies the entire width of the animal and in some instances may even distend the integument noticeably (FIG. 6, *K*, *b*; FIG. 7, *G*). The globose infective body, as in many other nematode-capturing hyphomycetes, disables the animal, and thereupon puts forth assimilative hyphae (FIG. 6, *H*, *J*; FIG. 7, *E*, *H*) which invade the helpless captive lengthwise (FIG. 6, *F*) until it is occupied from head to tail (FIG. 6, *I*, *K*; FIG. 7, *D*). Where the captured eelworm is very slender, like the eelworms that abundantly infested the plate cultures planted with the Oregon wheat straw (FIG. 6, *G–K*; FIG. 7, *D*, *F–H*), only a single assimilative hypha may be developed in most portions of the fleshy body, though stouter animals usually become permeated

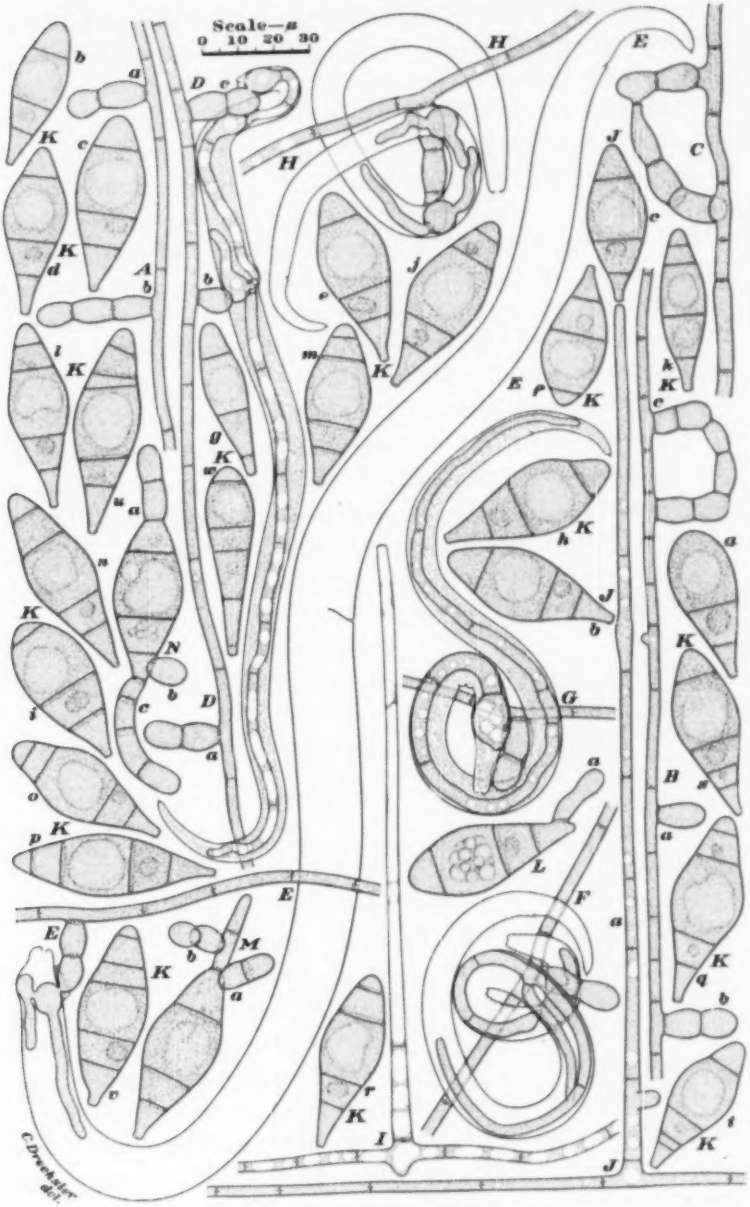

FIG. 7. *Dactylella cionopaga.*

by assimilative hyphae in numbers from two to six. Assimilative filaments of separate origin, and in stout eelworms assimilative hyphae arising from the same infective body, often become united distally through vegetative fusion (FIG. 6, *K*, *c*). After they have largely absorbed the globulose degenerating contents of the nematode, the assimilative hyphae show an increasingly vacuolate condition (FIG. 7, *D*). Ultimately as the animal becomes wholly depleted of digestible material the assimilative hyphae, through continued withdrawal of protoplasm backward into the external mycelium, are emptied of all living contents, and their membranous envelopes, together with the collapsing integument of the eelworm, gradually vanish from sight.

The fungus sporulates readily both in pure culture and on nematode-infested substratum. Its erect conidiophores (FIG. 6, *L–N*; FIG. 7, *I*) commonly vary from 170 to 300 μ in height, and thus seem generally shorter than those of *Dactylella gephyropaga*, as well as of most other similarly robust nematode-capturing hyphomycetes. Often a conidiophore (FIG. 7, *J*, *a*) after forming one conidium terminally (FIG. 7, *J*, *b*) will resume growth to produce a second conidium (FIG. 7, *J*, *c*) on a new tip farther upward; the first conidium being thereby pushed sideways and then presenting much the appearance of having been formed laterally. Occasionally growth is resumed once again, with ensuing development of a third conidium above the second, which in the meantime has like the first been brought into a lateral position.

The conidia thus formed, while of about the same width as those of *Dactylella gephyropaga*, are of noticeably greater length, and consequently have a somewhat more elongated shape together with greater volume. At the distal end they usually are broadly rounded, whereas proximally they taper toward a bluntly truncate base; so that they show various intergradations from a broadly fusiform to a clavate shape. In their septation they vary more than the conidia of *D. gephyropaga*, or of *D. aphrobrocha* and *D. bembicodes* among related constricting species. Many of the smaller, usually clavate specimens contain only two cross-walls (FIG. 6, *O*, *a*; FIG. 7, *K*, *a*). Among well developed conidia individuals with three (FIG. 6, *O*, *b–f*; FIG. 7, *K*, *b–i*) or four (FIG. 6, *O*, *g–n*; FIG. 7, *K*, *j–t*) cross-walls predominate, though not infrequently a five-

septate condition (FIG. 6, *O*, *o*, *p*; FIG. 7, *K*, *u–w*), virtually unknown in *D. gephyropaga* as also in *D. aphrobrocha*, and even a six-septate condition (FIG. 6, *O*, *q*) is encountered here. In quadriseptate specimens the cross-walls most commonly are placed in symmetrical arrangement (FIG. 6, *O*, *g*, *j*, *k*, *l*; FIG. 7, *K*, *j–r*) with a large cell occupying a median position between two smaller distal (apical and penultimate) cells and two smaller proximal (basal and parabasal) cells, yet rather often they occur in unsymmetrical arrangements with the large cell placed in penultimate (FIG. 6, *O*, *h*, *i*; FIG. 7, *K*, *s*, *t*) or in terminal (FIG. 6, *O*, *n*) position. The large cell of mature conidia contains usually a big conspicuous vacuole, while one or two of the remaining cells reveal commonly a globose mass that seems to be composed of densely conglomerated coarse granules. In some conidia where the large cell apparently begins to undergo division after its big vacuole has been formed, the belated cross-wall is laid down only within the parietal protoplasmic layer, leaving a large opening through which the vacuole extends from one daughter cell to the other (FIG. 6, *O*, *c*, *m*). Here and there a septum may be found that does not extend entirely across the conidium, but cuts off merely a small wedge-like portion from the proximal or the distal end of one of the larger cells (FIG. 6, *O*, *q*.)

Instead of germinating in commonplace manner by emission of a germ hypha from each end, many conidia after falling on a moist substratum give rise directly to adhesive outgrowths. Most frequently these outgrowths are put forth from one or from both of the end cells (FIG. 6, *P*, *a*; *Q*, *a*, *b*; *R*, *a*, *b*; *S*, *a*, *c*; *T*, *a*. FIG. 7, *L*, *a*; *M*, *a*; *N*, *a–c*), though sometimes they are found arising from intermediate cells (FIG. 6, *S*, *b*). Often, again, a conidium gives rise to one or more adhesive outgrowths (FIG. 6, *S*, *d*, *e*; *T*, *b*, *c*. FIG. 7, *M*, *b*) on a stout germ hypha. Such germ hyphae, which in themselves seem to lack adhesiveness, consist as a rule of only a few rather short segments, yet occasionally they may attain a length in excess of 100 μ (FIG. 6, *T*). While the adhesive outgrowths produced by detached conidia, like those arising from mycelial hyphae, commonly consist of one or two cells, in more than a few instances they are composed of three or four segments (FIG. 7, *N*, *c*).

In view of the frequently columnar shape of the predacious outgrowths a term compounded of two words meaning "pillar" and "trap," respectively, is proposed as specific epithet for the fungus.

Dactylella cionopaga sp. nov.

Mycelium sparsum; hyphis incoloratis, ramosis, mediocriter septatis, plerumque 2–5 μ crassis, hic illic prominentia glutinosa ferentibus; prominentibus glutinosis quandoque tuberiformibus sed saepius columnaribus, simplicibus vel parce ramosis, plerumque 10–90 μ longis, 5.5–10.5 μ crassis, in 1–7 (saepius in 1 vel 2) cellulis protoplasmate dense granuloso farctis consistentibus, ad septa leviter constrictis, quandoque inter se inordinatim conjunctis, rarius in laqueos quadrilateros vel semicirculos connexis—his prominentibus laqueisque vermiculos nematoideos errantes tenentibus, deinde tum integumentum animalis captivi perforantibus, tuber mortiferum intrudentibus, hyphas assumentes evolventibus quae carnem exhauriunt; hyphis assumentibus plerumque 2–5 μ crassis, mediocriter septatis. Hyphae fertiles incoloratae, erectae, vulgo simplices, 2–11 septatae, plerumque 170–300 μ altae, basi 5–7 μ crassae, sursum leniter attenuatae, apice saepe circa 3 μ crassae, ibi unum conidium ferentes, interdum recrescentes et 1 vel 2 alia conidia gignentes; conidiis incoloratis, plerumque late fusiformibus vel aliquid clavatis, apice late rotundatis, basi truncatis, vulgo 35–60 μ longis, 13–21 μ latis, 2–6 septatis plerumque triseptatis et quadriseptatis, post disjunctionem prominentia glutinosa praecipue ex apice et ex basi atque ex hypha germinationis brevi saepe emittentibus.

Vermiculos nematoideos diversos capiens consumensque habitat in stramento Tritici aestivi putrescenti prope Hermiston, Oregon, atque in humo silvestri prope Butternut, Wisconsin, et prope Presque Isle, Maine.

Mycelium scanty, spreading; vegetative hyphae colorless, septate, mostly 2 to 5 μ wide, here and there giving rise to adhesive outgrowths; adhesive outgrowths sometimes tuberiform but more often columnar, simple or branched, mostly 10 to 90 μ long, 5.5 to 10.5 μ wide, composed of 1 to 7 (usually of 1 or 2) cells densely filled with finely granular protoplasm, noticeably constricted at the septa, sometimes becoming fused with one another and thereby forming meshes which occasionally are rectangular or semicircular but more usually are of irregular shape—these outgrowths and meshes capturing nematodes through adhesion, later narrowly perforating the integument of each captured animal, then intruding a globose infective body and extending assimilative hyphae lengthwise through the interior to appropriate the fleshy contents; the assimilative hyphae 2 to 5 μ wide, septate at moderate intervals. Conidiophores colorless, erect, commonly unbranched, eventually divided by 2 to 11 cross-walls, mostly 170 to 300 μ high, 5 to 7 μ wide at the base, gradually tapering upward, about 3 μ wide at the tip, there producing a single conidium, though sometimes after repeated elongation forming one or two additional conidia. Conidia

colorless, mostly broadly spindle-shaped or somewhat clavate, broadly rounded at the distal end but at the narrower proximal end tapering toward the bluntly truncate base, commonly 35 to 60 μ long, 13 to 21 μ wide, containing 2 to 6 cross-walls though most usually triseptate or quadriseptate, often germinating by putting forth adhesive outgrowths especially from the basal and apical segments, though sometimes from an intermediate segment or from a short non-adhesive germ hypha.

Capturing and consuming different species of nematodes it occurs in decaying culms of *Triticum aestivum* near Hermiston, Oregon, and also in leaf mold from deciduous woods near Butternut, Wisconsin, and near Presque Isle, Maine.

A RETIARY DACTYLARIA PRODUCING TRISEPTATE CONIDIA FREQUENTLY OF UNUSUAL WIDTH

Agar plate cultures prepared, as opportunity offered, with small quantities of decaying material found on plant specimens received from Florida and Louisiana during the last 15 years have brought to light many of the nematode-capturing fungi that occur everywhere in the region surrounding Washington, D. C. In addition to *Arthrobotrys oligospora*, *A. conoides*, and *A. musiformis* Drechsler (**12**: 477–482) among the hyphomycetes employing adhesive networks for capturing eelworms, these plate cultures often displayed abundant development of predacious and reproductive apparatus wholly typical of *Dactylaria thaumasia*. Now and then they showed, further, a retiary form that arrested attention because many of its broad triseptate conidia appeared to be considerably wider than those of *D. thaumasia*. As the plate cultures were nearly always overrun by mites at an early stage, owing to heavy infestation of these animals in material from southern localities, they gave little encouragement for closer study or for isolation of the aberrant form. Better conditions for appropriate treatment were eventually given when presumably the same fungus developed rather extensively in several maizemeal-agar plate cultures which after being overgrown with mycelium of *Pythium ultimum* Trow had been further planted by adding small quantities of leaf mold gathered in deciduous woods near Roanoke, Virginia, on October 11, 1946. From observations on both pure and nematode-infested cultures the

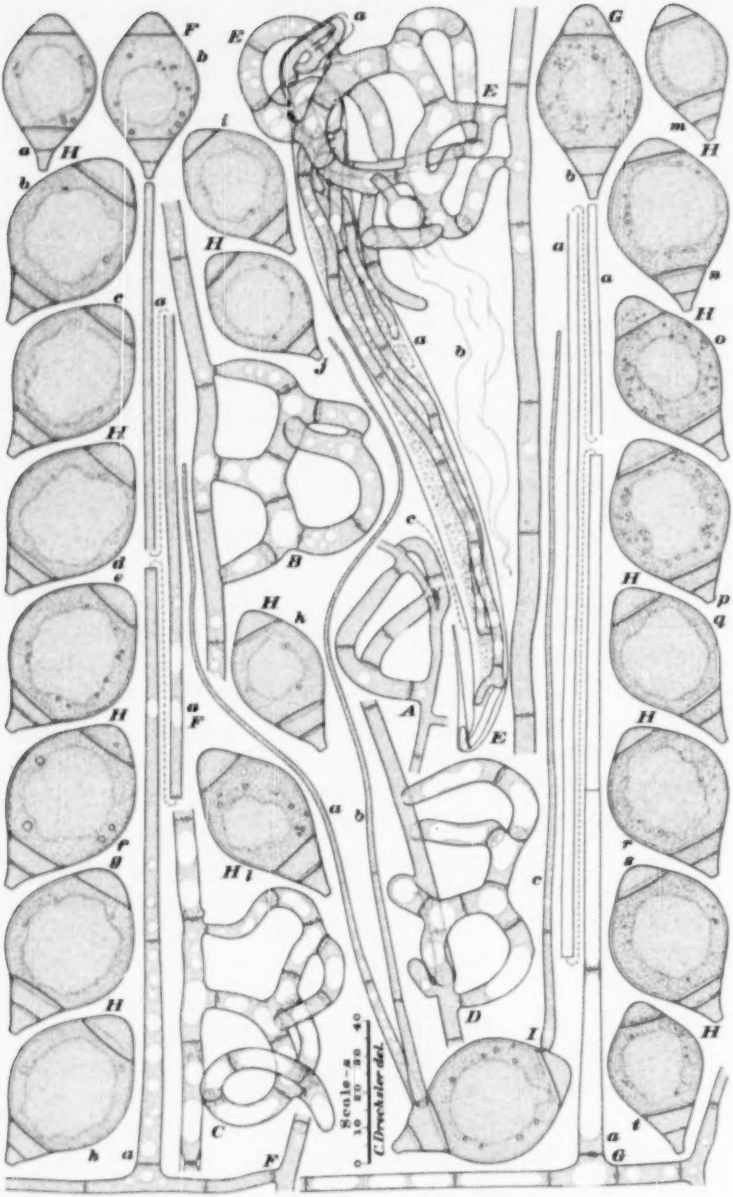

FIG. 8. *Dactylaria eudermata.*

fungus here concerned would seem to differ sufficiently from *D. thaumasia* to merit recognition as a separate species.

When sizable portions of agar well permeated with young mycelium are excised from pure cultures of the new hyphomycete and placed on agar plate cultures well infested with eelworms of such genera as *Rhabditis, Panagrolaimus, Acrobeloides,* and *Plectus,* new hyphae are extended in all directions from the transferred mycelium into the surrounding substratum. These radiating hyphae give rise at varying intervals to three-dimensional networks (FIG. 8, *A–D;* FIG. 9, *A, B*) which like the similar networks in many allied species are produced through successive development of rather thick recurving branches (FIG. 9, *C, a*). Each of these branches forms a closed semicircular loop or bail-like element by fusing terminally, at some distance from its origin, with a small protuberance put forth by a neighboring hyphal segment. The single fusion here takes place in much the same manner as each of the two fusions usually accomplished in the formation of a constricting ring. After the tips of the recurved branch and the opposed protuberance have come together somewhat broadly (FIG. 9, *C, b*) the portions of membrane at the surface of contact dissolve away (FIG. 9, *C, c*), though some time later the resulting continuity is modified by deposition of a cross-wall near the place of union (FIG. 9, *C, d*).

The hyphal bails and the networks compounded from them operate much like the similar apparatus employed by various other nematode-capturing hyphomycetes. Eelworms that in their continuous movement happen to run afoul of these structures are held fast despite their energetic struggles to escape. In some instances where little hyphal enwrapment is to be seen (FIG. 10, *A*) the captured animal must obviously be held mainly if not wholly through adhesion; but in other instances encirclement of the animal in one or several meshes (FIG. 9, *D*) gives reason for belief that it is held through both adhesion and entanglement. Following narrow perforation of the integument, the fungus disables the struggling eelworm by intruding one (FIG. 10, *A*) or more (FIG. 9, *D*) globose infective bodies from which assimilative hyphae are then extended (FIG. 9, *D*) to invade the fleshy interior from head to tail (FIG. 10, *B*). At first the assimilative hyphae are badly ob-

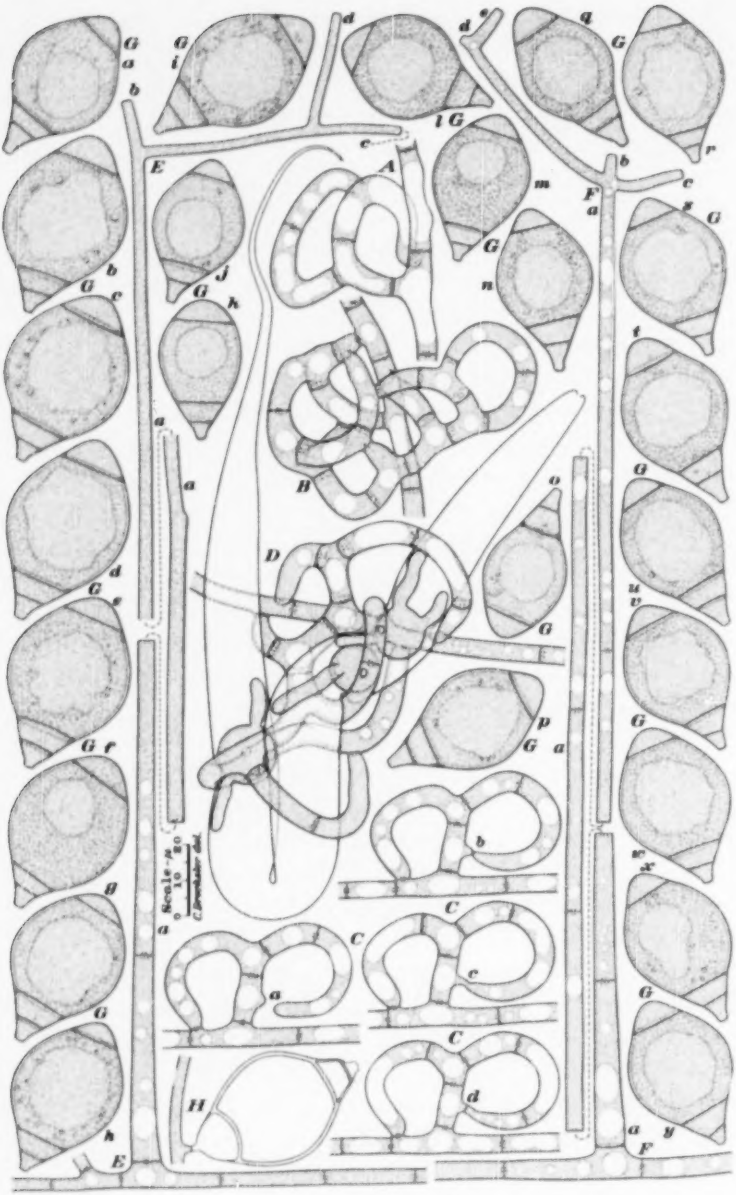

FIG. 9. *Dactylaria eudermata.*

scured from view owing to the globulose degeneration of the animal's musculature and organs. Later when the degenerating material has been largely absorbed (FIG. 8, *E, a*) the assimilative hyphae become more clearly visible, and instances of vegetative fusion between them are revealed (FIG. 8, *E, c*). At about this time they begin to show vacuoles. With further depletion of the digestible substance in the animal the vacuoles increase steadily in volume. Eventually the assimilative hyphae are wholly emptied of contents through withdrawal of their protoplasm backward into the external mycelium and, together with the collapsing pellicle, vanish from sight. In some instances the somewhat indurated infective body (FIG. 8, *E, b*) remains visible for a time after the associated hyphae have disappeared.

With ample nourishment being obtained through destruction of many nematodes the fungus soon produces conidiophores and conidia. The conidiophores arise in scattered positions from procumbent hyphae. Often they consist individually of a stout simple erect hypha, tapering gradually upward from base to tip, and ranging mostly from 0.4 to 0.5 mm. in height (FIG. 8, *F, a; G, a.* FIG. 10, *C, a*). These simple conidiophores bear terminally a single conidium (FIG. 8, *F, b; G, b.* FIG. 10, *C, b*) much like the simple conidiophores, for example, of *Dactylella aphrobrocha* and *D. bembicodes*, which, indeed, in their dimensions they resemble rather closely. The fungus likewise puts forth distally branched conidiophores (FIG. 9, *E, a; F, a*) that besides producing a spore at the tip of the main hypha (FIG. 9, *E, b; F, b*) bear additional spores singly on its primary branches (FIG. 9, *E, c; F, c, d*) and also on its secondary branches (FIG. 9, *E, d; F, e*) if such are present. As the branches are often rangy, sometimes exceeding 50 μ in length, the conidia produced plurally are in many instances attached at generous distances from one another. Nevertheless where conidia are held aloft in numbers of three or four they usually offer a clustered aspect, for owing to the large size of the individual spores the intervening spaces do not seem disproportionately wide.

The conidia produced by the fungus in nematode-infested culture (FIG. 8, *H, a–t;* FIG. 9, *G, a–y;* FIG. 10, *D, a–f*) owe their frequently large size more to their unusual width than to their length.

On the whole they appear about one-third wider and one-sixth longer than the similarly turbinate triseptate conidia of *Dactylaria thaumasia*. In fully developed spores the triseptate condition is so strongly predominant that the few biseptate specimens (FIG. 10, *D, f*) commonly present arouse misgivings as to their maturity, while quadriseptate conidia seem scarcely less alien to the fungus than to *Dactylella stenobrocha* and *Dactylella bembicodes*. As in *D. bembicodes* the three cross-walls are usually so placed that the penultimate cell greatly exceeds the others in dimensions and volume. This voluminous cell contains a large irregular vacuole, and is further distinguished by being surrounded with a noticeably thickened wall. When nematode-infested cultures, on being uncovered for microscopical examination, are exposed to dry air, the thin-walled apical cell collapses badly, whereas the sturdy penultimate cell, at least in respect to its peripheral outer membrane, commonly retains its shape; the result being that the conidia held aloft offer a curiously truncated appearance. Detached conidia are often found bearing one or more slender filamentous aerial outgrowths (FIG. 8, *I, a–c*), sometimes over 200 μ long, which originate commonly from the parabasal and apical cells. Under somewhat humid and rather warm (25° to 35° C.) conditions such outgrowths are extended freely also from conidia still supported on the conidiophores. In water or on a moist nutrient substratum germination takes place by the emission of one or more submerged germ hyphae (FIG. 10, *E*). Fusion of germ hyphae with mycelial filaments is frequent and often leads to evacuation of all protoplasmic contents from the conidia concerned (FIG. 9, *H*; FIG. 10, *F*).

The fungus grows rapidly in pure culture on maizemeal agar. In test-tube cultures on this substratum it sometimes produces a more copious aerial mycelium than I have hitherto observed in cultures of any other nematode-capturing hyphomycete. Often the column of rather loose cottony growth immediately above an agar slant is surmounted by an upper layer of firm felt-like texture; the appearance given then being rather similar to that commonly presented by tube cultures of my *Pythium arrhenomanes*. A few days after a pure culture has been planted it begins to produce conidia, which for the most part, however, reveal only

rather poorly the morphological features distinctive of conidia formed on nematode-infested substratum. Occasional specimens, it is true, are divided by three cross-walls into four cells whereof the one in penultimate position exceeds the others in size, but even in these specimens departure from the normal is often clearly evident in greatly reduced size of the apical and basal segments (FIG. 10, *G*, *a–c*). Very frequently the conidia formed in pure culture contain only two cross-walls, both of them placed near the proximal end, so that the large globose cell in terminal position here surmounts the much smaller parabasal and basal segments (FIG. 10, *G*, *d–m*). Frequently, again, the conidia contain only a single cross-wall, which being placed near the basal end delimits a large globose distal cell from a small proximal cell (FIG 10, *G*, *n–q*). Like the spores formed in nematode-infested culture those produced in pure culture are often found bearing one or more aerial outgrowths of slender filamentous shape (FIG. 10, *H*), or are provided with broader germ hyphae of ordinary vegetative character (FIG. 10, *I–K*). The germ hyphae here likewise often fuse with neighboring mycelial filaments (FIG. 10, *L*, *M*), thereby establishing a passageway through which the protoplasmic materials of the conidium may migrate into the mycelium.

Despite their frequently rather different appearance the conidia produced by the fungus in pure culture are not essentially different in kind from the conidia formed on nematode-infested substratum. They are manifestly not referable to a separate category of spores, such as is recognizable in the uniseptate allantoid conidia of *Dactylella heterospora* and in the globuliferous conidioid bodies of *D. doedycoides*. Their generally smaller size, the disproportionate reduction of their smaller cells, and more especially the usual occurrence of their large cell in distal position owing to frequent lack of a small cell at the apex, seem to come about as modifications resulting from the different nourishment supplied in maize-meal agar not inhabited by eelworms. It seems possible that the modifications in question fall outside the range of ordinary variation, and perhaps are to be construed as pathological abnormalities deriving from nutritional deficiency. In any case the tendency toward morphological deterioration here offers marked contrast

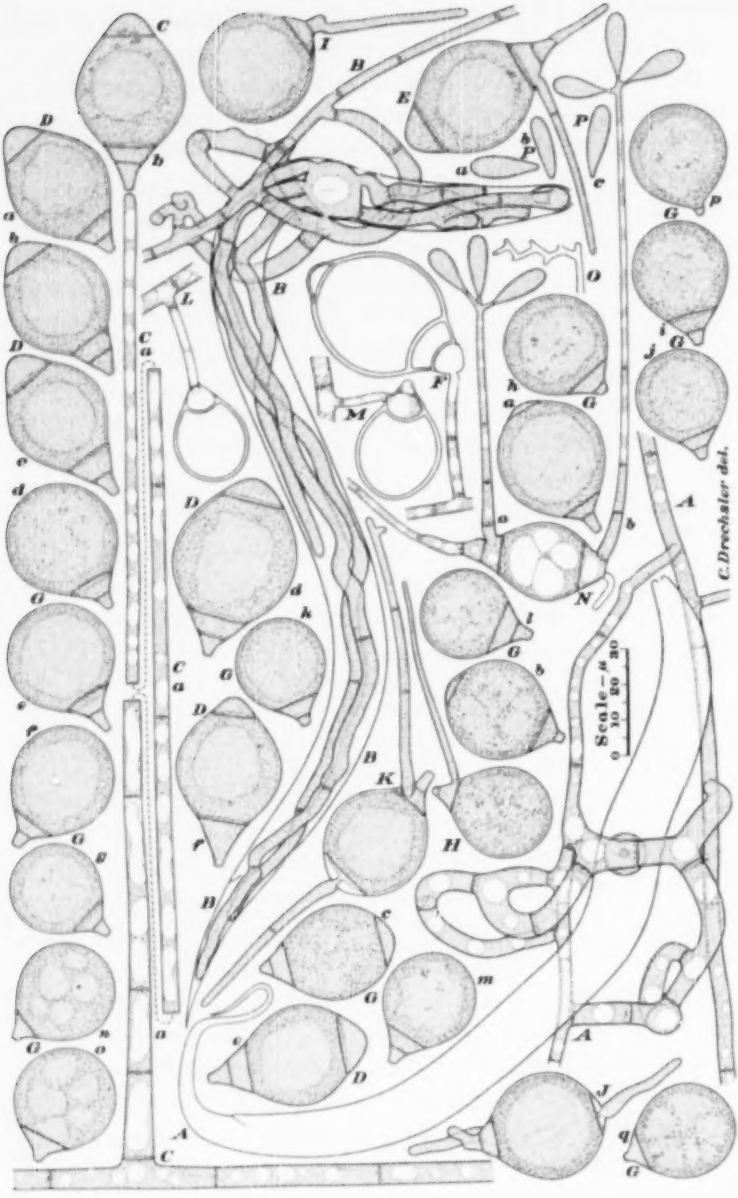

FIG. 10. *Dactylaria eudermata.*

with the luxuriant and wholly correct sporulation of *Dactylaria thaumasia* when grown in pure culture on maizemeal agar.

Yet conidia essentially different in kind from the broad triseptate spores hitherto discussed have come under observation. One of the original plate cultures in which the fungus had grown out directly from plantings of leaf mold showed within a limited area a number of detached triseptate conidia that after falling on the moist agar substratum had put forth one or more germ conidiophores (FIG. 10, *N*, *a*, *b*) bearing small secondary conidia in loose capitate arrangement. These germ conidiophores, measuring 65 to 275 μ in height and 3 to 4 μ in basal diameter, tapered gradually upward to a width of 1.5 to 2 μ at the tip, where they terminated usually in a few—mostly in 2 to 5—sterigmatic spurs each bearing a secondary conidium; though in some instances secondary conidia up to seven in number were found borne on spurs, each about 5 μ long, produced one after another by successive subapical branching (FIG. 10, *O*). The secondary conidia (FIG. 10, *P*, *a–c*) were colorless, unseptate, of clavate shape, 15 to 25 μ long, and 4.3 to 6.5 μ wide. In the same area of the Petri plate culture similar conidia were borne also on small conidiophores arising from mycelial filaments. Efforts to start pure cultures from the meager supply of unseptate conidia were unsuccessful, and the cultures started from broad triseptate conidia taken from tall conidiophores in the area have not so far yielded a secondary or regularly unicellular conidial stage. Although it seems probable that the subsidiary spore stage belonged in the life history of the present fungus, the connection has not yet been established. Much the same uncertainty remains here as was noted earlier (**16**: 163–166) in respect to the likely connection of a rather similar secondary conidial stage with *Dactylaria psychrophila*.

The fungus offers further parallelism with *Dactylaria psychrophila* in having a sporulating habit so meagerly capitate that its assignment to the genus *Dactylaria* needs to be justified in part by its manifestly close relationship to the more pronouncedly capitate *D. thaumasia*. As its adhesive meshes are of about the same dimensions as those of *D. thaumasia* and most other retiary hyphomycetes, its networks appear not to share the somewhat closer texture noted in the predacious apparatus of *D. psychro-*

phila; and certainly they do not share the markedly closer texture usual in networks of my *D. polycephala* (**12**: 527–531). Like *D. psychrophila* the fungus produces larger conidia that *D. thaumasia,* but its conidia owe their larger size mainly to greater width, whereas those of *D. psychrophila* owe their larger size mainly to greater length. Quadriseptate partitioning, so infrequent in conidia of the fungus, is commonplace among conidia of *D. psychrophila.* Pure cultures of the fungus on maizemeal agar have not so far shown chlamydospores or any other kind of indurated bodies; whereas similar cultures of *D. thaumasia* commonly produce thick-walled chlamydospores in such abundance that the substratum is given a reddish coloration.

A term meaning "with good, stout hide" may serve conveniently as specific epithet for the new hyphomycete in recalling the thick membrane surrounding the massive penultimate segment of its conidia.

Dactylaria eudermata sp. nov.

Mycelium effusum; hyphis sterilibus incoloratis, mediocriter septatis, plerumque 1.8–7.5 μ crassis, laqueos tenaces arcuatos vel circulares in reticula saepe conjunctos proferentibus; his laqueis reticulisque vermiculos nematoideos illaqueantibus, deinde tum integumentum animalis captivi anguste perforantibus, tuber mortiferum globosum intrudentibus, hyphas intus evolventibus quae carnem exhauriunt. Hyphae fertiles incoloratae, erectae, plerumque 2–8 septatae, vulgo 400–500 μ altae, basi saepius 6–9 μ crassae, sursum leniter attenuatae, apice 2.5–3.5 μ crassae, simplices vel prope apicem aliquid ramosae, itaque nunc unicum conidium gignentes, nunc 2–4 conidia in capitulum laxum ferentes; conidiis incoloratis, ellipsoideis vel obovoideis vel late turbineis, apice rotundatis, deorsum paulum attenuatis, basi truncatis, plerumque 37–55 μ (saepe circa 48 μ) longis, 21–35 μ (saepe circa 28 μ) crassis, vulgo triseptatis, paenultima eorum cellula 21–35 μ (saepe circa 29 μ) longa, aliis cellulis eorum multo minoribus, inter se subaequalibus, vulgo 4–8 μ (saepe circa 6 μ) longis; interdum omnibus cellulis glabris, interdum aliquibus cellulis minoribus 1 vel 2 appendicibus praeditis; appendicibus incoloratis, filiformibus, rectis vel flexuosis, continuis vel prope basim 1–2 septatis, plerumque 50–275 μ longis, basi 2–4 μ crassis, sursum leniter attenuatis, apice circa 1 μ crassis.

Vermiculos nematoideos diversos capiens consumensque habitat in humo silvestri prope Roanoke, Virginia.

Mycelium spreading; vegetative hyphae colorless, septate at moderate intervals, mostly 1.8 to 7.5 μ wide, often especially in the presence of nematodes giving rise to arched or circular hyphal meshes, which, though at first discrete, are later frequently com-

pounded into more or less extensive networks; the meshes and networks capturing nematodes through adhesion and entanglement, then narrowly perforating the integument of each captured animal and intruding one or more infective bodies of subspherical shape, from which are extended assimilative hyphae, mostly 2 to 5 μ wide, to appropriate the fleshy contents. Conidiophores colorless, erect, usually containing 2 to 8 cross-walls, commonly 400 to 500 μ high, 6 to 9 μ wide at the base, tapering gradually upward to a width of 2.5 to 3.5 μ near the tip, simple or somewhat branched at the distal end, consequently sometimes producing a single conidium and at other times bearing 2 to 4 conidia in loose capitate arrangement. Conidia colorless, ellipsoidal or obovoid or broadly turbinate, broadly rounded at the tip, somewhat tapered proximally, truncate at the base, mostly 37 to 55 μ (average about 48 μ) long, 21 to 35 μ (average about 28 μ) wide, commonly triseptate, the penultimate cell 21 to 35 μ (average about 29 μ) long, the other three cells much smaller, individually most often 4 to 8 μ (average about 6 μ) long and sometimes bearing 1 or 2 aerial outgrowths; aerial outgrowths colorless, filamentous, straight or flexuous, continuous or with 1 or 2 cross-walls in the proximal portion, 50 to 275 μ long, 2 to 4 μ wide at the base, tapering gradually upward to a width of approximately 1 μ at the tip.

Capturing and consuming nematodes of different species, it occurs in leaf mold in deciduous woods near Roanoke, Virginia.

A CANDIDA-LIKE DACTYLARIA CAPTURING EELWORMS BY ADHESION TO SLENDER-STALKED ADHESIVE KNOBS

Several maizemeal-agar plate cultures which after being permeated with mycelia of *Pythium mamillatum* Meurs had been further planted on April 29, 1948, with small quantities of partly decayed bluegrass (*Poa pratensis* L.) detritus newly collected in a field near Beltsville, Maryland, showed 16 days later some expanse of conidial apparatus rather similar to that of *Dactylaria candida* (Nees) Sacc., but differing in being less strongly capitate. A more decisive difference came to light when the underlying mycelium was examined, for though the vegetative hyphae manifestly obtained their nourishment by capture of nematodes abundantly infesting the substratum, the animals were in all instances held fast through adhesion to stalked globose unicellular knobs comparable in size and efficacy to the adhesive knobs utilized by my *Dactylella*

asthenopaga (12: 496–499) and my *Dactylaria haptospora* (13: 456–461) as well as by the nematode-capturing fungus (12: 492–496) held referable to *Dactylella ellipsospora* Grove (22). The mycelium was wholly lacking in non-constricting rings, such as those employed very effectively by the widespread nematode-capturing hyphomycete I have assigned (12: 523–527) to the ancient species of Nees von Esenbeck. However, since this hyphomycete, besides producing non-constricting rings, gives rise terminally on very slender stalks to small globose unicellular knobs which in soft agar cultures have only rarely been found operative in capturing eelworms, it was necessary to consider whether the observed absence of non-constricting rings together with the greater size and efficiency of the globose knobs might perhaps have resulted from unusual environal conditions. The fungus growing out from the bluegrass detritus was therefore isolated by removing its conidia from the erect conidiophores to sterile agar. From the pure cultures thus obtained portions of agar well permeated with young mycelium were excised and placed on Petri plate cultures that had become infested with saprophilous nematodes introduced on decaying materials of various kinds. Among the cultures used in these trials were some that had been started by planting the sterile substratum with pieces of softened discolored roots of matai or Chinese waternut [*Eleocharis dulcis* (Burm. f.) Henschel] plants received from Winter Park, Florida, late in July, 1948. After being promptly overgrown by my *Pythium myriotylum* the agar here afforded abundant multiplication of a slender species of *Panagrolaimus*, which later suffered destruction in large numbers when young mycelium of the Beltsville hyphomycete was superadded. As the invasive development of predacious and parasitic fungi is in general more clearly visible in slender than in stout eelworms, material pertaining to or resulting from the destruction of the *Panagrolaimus* was used advantageously in preparing the figures relating to the fungus (FIGS. 11, 12).

None of the nematode-infested cultures in which portions of young mycelium were planted showed any development of non-constricting rings. From the superadded mycelium rather slender filaments grew out that at somewhat variable intervals and mostly in submerged positions consistently gave rise to unicellular adhesive

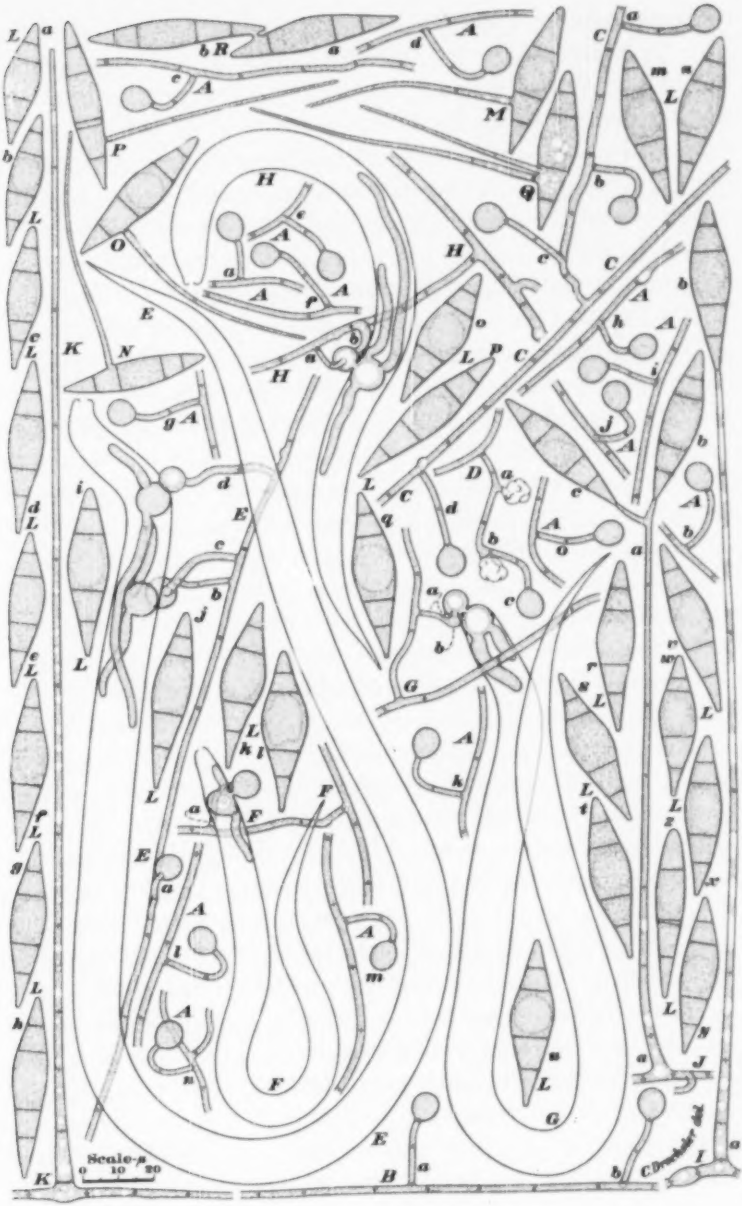

FIG. 11. *Dactylaria haptotyla.*

knobs on stalks sometimes consisting of one cell (FIG. 11, *A*, *a*, *b*. FIG. 12, *A*, *a–d*; *D*, *a*) but more often consisting of two cells (FIG. 11, *A*, *c–o*; *B*, *a*, *b*; *C*, *a–d*; *E*, *a*. FIG. 12, *A*, *e–k*; *B*, *a*, *b*; *C*, *b*) and occasionally consisting of three cells (FIG. 12, *C*, *c*). Most frequently the stalks were 1.4 to 1.9 μ wide and from 10 to 25 μ long. They appear appreciably stouter, therefore, than the corresponding stalks of *Dactylaria candida*, which, as a rule, measure only 1 to 1.4 μ in width. At the same time they seem markedly more slender as well as generally longer than the homologous stalks of *Dactylella ellipsospora*, since these usually measure 2.4 to 3 μ in width and 5 to 10 μ in length; the dimensional contrast here offering some parallelism with the contrast between the stalks supporting the constricting rings of *Dactylella aphrobrocha* and those supporting the constricting rings of *Dactylella bembicodes*. Nearly the same degree of contrast is evident when comparison is extended to the stalks of *Dactylella asthenopaga*, which, as is apparent from their recorded dimensional ranges—2 to 3 μ for width, and 3 to 10 μ for length—are similarly of short sturdy conformation. The stalks of *Dactylaria haptospora*, commonly 1.5 to 2.5 μ wide and 4 to 30 μ long, offer notably better agreement with respect to measurements.

The globose or prolate ellipsoidal adhesive knobs of the Beltsville fungus have been found varying mostly from 7 to 10 μ in length and from 6 to 8.5 μ in width. As has been intimated they differ little in size from the adhesive knobs of the three previously described nematode-capturing hyphomycetes that produce no other type of predacious organ; though on a strict comparison of relevant measurements they would seem slightly smaller than the knobs of *Dactylella ellipsospora*, and slightly larger than those of *Dactylella asthenopaga* and *Dactylella haptospora*. They are markedly larger, of course, than the meagerly operative knobs of *Dactylaria candida*, which commonly measure only 4 to 7 μ in length and 3.8 to 6 μ in width. In nearly equal measure they exceed in size also the meagerly operative knobs of my *Dactylella lysipaga* that usually range from 5 to 8 μ in length, and from 4.5 to 6 μ in width.

Frequently in predacious organs that have attained some age without capturing a nematode, the adhesive cell loses its protoplasmic contents and collapses emptily (FIG. 11, *D*, *a*) as the supporting stalk grows out obliquely near the distal end to form a sec-

ond adhesive cell on the prolongation. The same process may then be repeated, with the second adhesive cell in turn losing its contents and collapsing (FIG. 11, *D, b*) as a third adhesive cell (FIG. 11, *D, c*) is produced. In such successive elongation each new increment is at first continuous throughout. Indeed, young stalks generally (FIG. 12, *C, a*) remain in an unseptate condition until the terminal swelling destined to be delimited as an adhesive knob is nearly full grown.

The glandular knob, if left undisturbed, does not show under ordinary microscopical inspection any recognizable coating of adhesive material on its surface. An eelworm coming in contact with it is nevertheless held securely despite energetic struggles to escape. After these struggles have proceeded for some time, a cushion of colorless glutinous substance often becomes visible between the knob and the integument of the captured animal (FIG. 12, *D, b; E, a; F, a*). Soon the adhering knob perforates the integument by means of a slender protuberance, which, on reaching the fleshy interior, immediately widens out into a globose infective body that usually continues to expand (FIG. 12, *D*) until it extends entirely or almost entirely across the nematode (FIG. 11, *E–H;* FIG. 12, *E*). The nematode gradually becomes enfeebled from the injury thus sustained, and thereupon is invaded further by assimilative hyphae arising from the infective bodies (FIG. 11, *E–H;* FIG. 12, *E, F*). These assimilative hyphae grow lengthwise through the animal until they occupy it from head to tail (FIG. 12, *G, H*). At first, even in slender eelworms, they are to a considerable degree obscured from view, owing to the globulose degeneration of musculature and organs that accompanies their progress. Later, when the degenerating materials have been largely absorbed, they become more clearly visible. With further reduction of the animal's substance, they show an increasingly vacuolate condition (FIG. 12, *H*) as their protoplasmic contents are withdrawn backward into the external mycelium. Eventually they are completely emptied of living protoplasm, and their membranous envelopes, together with the collapsing integument surrounding them, disappear from sight.

In the withdrawal of protoplasmic contents from the assimilative hyphae into the external mycelium, the passageway provided by the narrow stalk originally supporting the adhesive knob (FIG. 11, *E, b;*

H, a. FIG. 12, *D, b; F, a*) is often supplemented by a passageway provided by a second and noticeably wider hyphal element (FIG. 11, *E, c; H, b.* FIG. 12, *D, c; F, b*) connecting the adhesive knob with the mycelial filament at a point a short distance from the origin of the stalk. This second hyphal connection is often present at an early stage in the invasion of the eelworm, and may possibly be formed to replace the stalk, which, owing to its slenderness, must often incur injury from the struggles of the animal. It may be presumed that localized injury occasions the disappearance of protoplasm within the distal portion of the stalk (FIG. 11, *G, a;* FIG. 12, *H, a*) in the rather frequent instances where the new hyphal element (FIG. 11, *G, b;* FIG. 12, *H, b*) is found arising from a median position in the stalk rather than from the parent mycelial filament. Sometimes the original stalk seems to disappear completely, so that the supplementary hyphal element, recognizable by its greater width, provides the only communication between the adhesive knob and the mycelium (FIG. 11, *E, d.* FIG. 12, *G, a; H, c*). On the other hand, in instances where the captured eelworm is of relatively small size, the adhesive knob may remain attached, at least during the earlier stages of invasion, solely by the stalk on which it was produced (FIG. 11, *F, a;* FIG. 12, *E, a*).

The fungus usually puts forth conidiophores and conidia in moderate abundance both in pure culture and on nematode-infested substratum. Very often, in the beginning, most of the conidiophores present in a stand (FIG. 11, *I, a; J, a; K*) will bear spores singly (FIG. 11, *I, b*) or in twos (FIG. 11, *J, b, c*). At this stage the fungus looks much like a *Dactylella*. Later, however, with the production of additional spores on short branches or spurs, many conidiophores (FIG. 12, *I, a, b; J*) come to bear three (FIG. 12, *I, c–e*) or four (FIG. 12, *I, f–i*) or as many as five conidia in loosely capitate arrangement, thereby acquiring an appearance and habit usually associated with the genus *Dactylaria*. On the whole, capitate development here would seem tardier and less pronounced than in *Dactylaria candida*.

In their handsome fusiform shape the conidia rather closely resemble those of *Dactylella ellipsospora*, *Dactylella lysipaga*, and *Dactylaria candida*, while differing markedly from the clavate spores of *Dactylella asthenopaga*, as well as from the elongated cylindrical

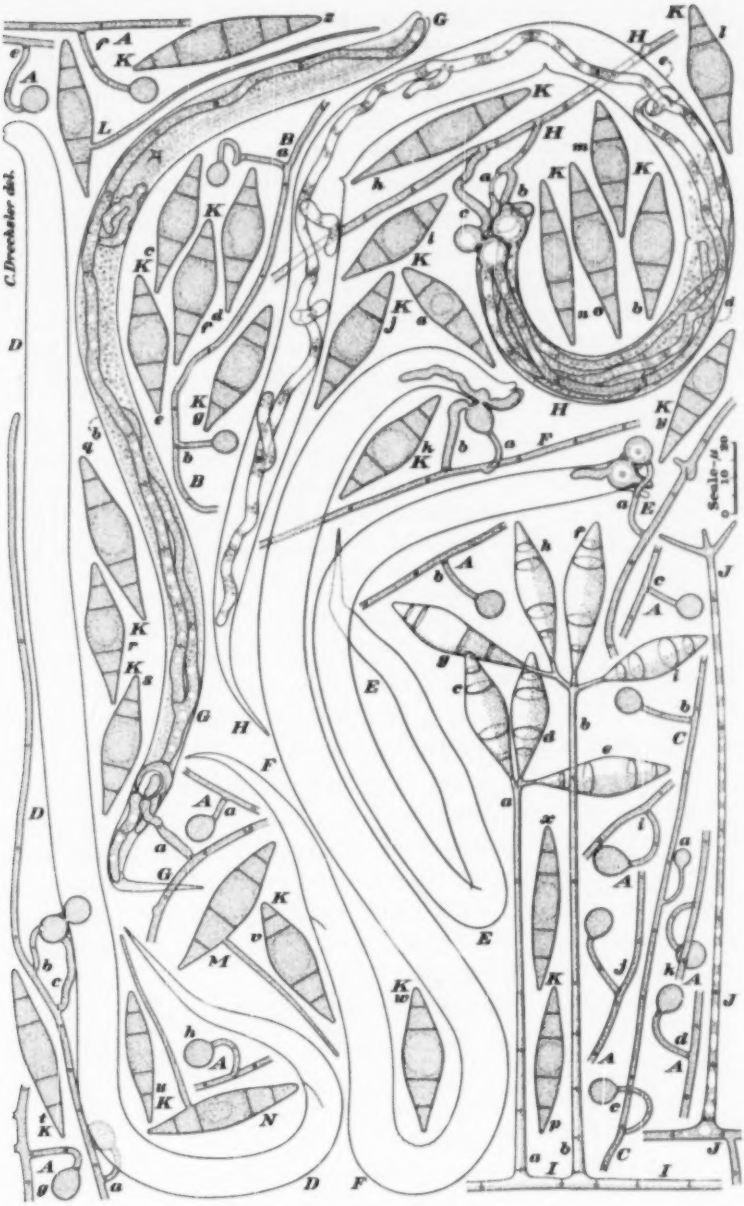

FIG. 12. *Dactylaria haptotyla.*

conidia of *Dactylaria haptospora*. When fully developed they contain three (FIG. 12, *K*, *a*, *b*) or four (FIG. 11, *L*, *a–z*; FIG. 12, *K*, *c–x*) or occasionally five (FIG. 11, *P*; FIG. 12, *K*, *y*, *z*) cross-walls. The quadriseptate condition, with the cross-walls so placed that a largish median cell is delimited between two smaller proximal cells and two smaller distal cells, usually predominates, and thus seems to represent the manner of septation most characteristic of the species. Conidia having such septation give measurements for length ranging from 34 to 55 μ, and measurements for width ranging from 7.4 to 13.3 μ; the computed averages for these dimensions being 43.7 μ and 10.7 μ, respectively. Computed values for average lengths of the five component cells are as follows: basal cell, 9 μ; parabasal cell, 7.3 μ; middle cell, 14.4 μ; penultimate cell, 6.1 μ; apical cell, 6.9 μ. Owing to the greater variability generally evident here in the size and septation of the conidia, the measurements just given must be held less accurately descriptive of the species than are the corresponding measurements, for example, of *Dactylella aphrobrocha*.

Often while it is still borne aloft on the conidiophore, or after it has fallen off, a conidium, whether triseptate (FIG. 11, *M*), quadriseptate (FIG. 11, *N*, *O*; FIG. 12, *L–N*), or quinqueseptate (FIG. 11, *P*), will extend a slender tapering filamentous outgrowth into the air. These outgrowths, which commonly are 25 to 100 μ long, 1.5 to 2.5 μ wide at the base, and about 1 μ wide near the tip, arise most usually from the parabasal segment. Sometimes a conidium will put forth two such aerial outgrowths (FIG. 11, *Q*). Two detached conidia (FIG. 11, *R*, *a*, *b*) lying near together on the surface of a culture often become fused to one another in much the same manner as neighboring mycelial filaments or neighboring assimilative hyphae (FIG. 12, *G*, *b*; *H*, *d*, *e*).

A term compounded of two words meaning "to fasten," and "knob," respectively, is deemed suitable as specific epithet for the fungus.

Dactylaria haptotyla sp. nov.

Mycelium sparsum; hyphis sterilibus incoloratis, mediocriter septatis, plerumque 1.4–3.8 μ crassis, bullas globosas vel ellipsoideas, 7–10 μ longas, 6–8.5 μ crassas, ex ramulo recto vel curvato, saepius 7–27 μ longo, 1.4–1.9 μ crasso, in 1–3 cellulis consistente, singillatim hic illic emittentibus; his bullis ad vermicu-

los nematoideos inhaerentibus, itaque animalia tenentibus, integumentum eorum perforantibus, tuber mortiferum intrudentibus, hyphas intus evolventibus quae carnem exhauriunt. Hyphae fertiles incoloratae, erectae, saepius 4–11 septatae, 115–325 μ altae, basi 3–4.5 μ crassae, sursum leniter attenuatae, apice circa 1.5 μ crassae, primum saepe simplices et in unum conidium abeuntes, deinde apice saepe parce ramosae denique 2–5 conidia in capitulum laxum ferentes; conidiis hyalinis, vulgo fusoideis, basi truncatis, apice anguste rotundis, plerumque 33–55 μ (saepe circa 43.7 μ) longis, 7.4–13.3 μ (saepe circa 10.7 μ) crassis, 3–5 septatis, saepissime quadriseptatis denique cellula antepaenultima crassiore et longiore quam aliis cellulis.

Vermiculos nematoideos specierum multarum capiens consumensque habitat in foliis caulibusque Poae pratensis putrescentibus prope Beltsville, Maryland.

Mycelium scanty; vegetative hyphae colorless, septate at moderate intervals, mostly 1.4 to 3.8 μ wide, often especially in the presence of nematodes, giving rise here and there on stalks frequently straight or somewhat curved, 7 to 27 μ long, 1.4 to 1.9 μ wide, sometimes unicellular but usually bicellular and occasionally tricellular, to unicellular knobs subspherical or prolate ellipsoidal in shape, 7 to 10 μ long and 6 to 8.5 μ wide; the knobs holding fast to nematodes, individually perforating the integument of the adhering animal, then intruding a globose infective body from which assimilative hyphae are extended to appropriate the fleshy contents. Conidiophores hyaline, erect, often containing 4 to 11 cross-walls, usually 115 to 325 μ high, 3 to 4.5 μ wide at the base, tapering gradually upward, about 1.5 μ wide at the tip, at first often simple and terminating in a single conidium, later often provided near the apex with a few short branches or spurs, and then bearing 2 to 5 conidia in loose capitate arrangement; conidia colorless, usually spindle-shaped, tapering downward toward the narrow truncate base, somewhat narrowly rounded at the distal end, mostly 33 to 55 μ (average about 43.7 μ) long, 7.4 to 13.3 μ (average about 10.7 μ) wide, containing 3 to 5 cross-walls but most often divided by 4 cross-walls into 5 cells whereof the one in middle position usually exceeds the others in length and width.

Capturing and consuming nematodes of different species it occurs in decaying leaves and stems of *Poa pratensis* near Beltsville, Maryland.

A CANDIDA-LIKE DACTYLARIA WITH EFFICACIOUS ADHESIVE KNOBS AND INDURATED HYPHAE

A nematode-capturing fungus which like *Dactylaria haptotyla* resembles *Dactylaria candida* in its conidial apparatus came to light in several maizemeal-agar plate cultures that after being

overgrown by my *Pythium arrhenomanes* had been further planted with small quantities of partly decayed vegetable refuse taken from a garden in Roanoke, Virginia, on October 11, 1946. In these cultures the massive lobulate sporangial complexes that were soon produced everywhere by *P. arrhenomanes* served as a very abundant food supply for stylet-bearing nematodes addicted to feeding on fungus protoplasm after the manner described by Christie and Arndt (**6**) for *Aphelenchoides parietinus* (Bastian **1865**) Steiner **1932**. Consequently the parasitic nematodes prospered greatly, and after two weeks were present in unwonted numbers. About 15 days after the vegetable detritus had been added the stylet-bearing animals, as well as the saprophilous eelworms intermingled with them, began to suffer noticeable losses from the destruction of many individuals by predacious mycelia that grew from the opaque material into the subjacent agar. Capture of nematodes was effected here exclusively by adhesive knobs of a size then known to me—*Dactylaria haptotyla* not having been discovered at the time—only in *Dactylella ellipsospora*, *Dactylella asthenopaga*, and *Dactylaria haptospora*. When, as soon happened, conidiophores arose from the predacious mycelia, they bore conidia which, while differing markedly from those of *Dactylella asthenopaga* and *Dactylaria haptospora*, resembled in a general way the conidia of *Dactylella ellipsospora* with respect to outward shape and to septation. Agreement with respect to sporulating habit was lacking, however, for the conidia here, instead of being borne for the most part singly, were held aloft plurally in loose heads, thereby offering much the same appearance as *Dactylaria candida*. To determine more especially whether non-constricting rings, familiar as the efficient predacious organs of *Dactylaria candida*, might be formed under changed conditions, the Roanoke fungus was isolated through removal of conidia from the loose heads to sterile agar; and from the pure cultures thus obtained sizable portions of its mycelium were cut out and placed on Petri plate cultures that had become well infested with nematodes originating from different kinds of decomposing plant materials. Development of non-constricting rings never ensued in these trials. The fungus, under varying conditions, continued to capture nematodes solely by means of adhesive knobs.

The adhesive knobs are borne on stalks most often, perhaps, consisting of two cells (FIG. 13, *A*, *a–q*; *B*, *a*; *C*, *a*, *b*; *D*, *a*; *E*, *a*, *b*), but nearly as often consisting of a single cell (FIG. 13, *C*, *c*; *D*, *b–e*; *F*; *G*, *a*; *H*, *a*; *K*, *a*). In stalks composed of more than one cell the basal segment may include a portion of the parent hypha (FIG. 13, *B*, *b*; *I*; *J*), or may even extend into a branch arising nearby from the parent hypha (FIG. 13, *K*, *b*). The stalks are commonly from 2 to 2.5 μ wide and from 10 to 20 μ long, though some unicellular examples do not greatly exceed 5 μ in length (FIG. 13, *D*, *b*; *G*, *a*), while among the four-celled specimens (FIG. 13, *L*) occasionally to be found some measure about 50 μ in this dimension. They would seem, therefore, somewhat wider than the stalks in *Dactylaria haptotyla*, and, to extend the comparison, are appreciably narrower and also longer than the stalks of *Dactylella ellipsospora*.

The knobs of the Roanoke hyphomycete, like the similar organs in allied species, show no visible coating of adhesive material as long as they are left undisturbed. Nevertheless, eelworms that come into contact with them are held fast despite prolonged struggles to escape. Some little time—frequently about 15 or 30 minutes—after capture of a nematode has been effected, a cushion of hyaline glutinous material can be seen between the knob and the animal's integument (FIG. 13, *D*, *d*). Soon the knob puts forth centrally in the region of contact a slender protuberance that penetrates the integument to give rise within to a globose infective body. This body usually expands until it occupies the entire width of the animal (FIG. 13, *D*, *d*; *E*, *c*, *d*; *G*, *b*, *c*; *H*, *b*; *M*; *N*; *O*), which thus is severed internally and as a result becomes incapable of energetic movement. Thereupon the infective body produces assimilative hyphae that continue to grow lengthwise through the fleshy interior (FIG. 13, *N*, *O*) until the eelworm is permeated from head to tail (FIG. 13, *D*, *d*; *E*, *c*, *d*; *G*, *b*, *c*; *H*, *b*; *M*). At first the assimilative hyphae are only indistinctly visible, being then obscured because of the globulose degeneration marking their progress through musculature and organs; though in slender nematodes, such as the stylet-bearing parasites found utilized as prey in the original cultures (FIG. 13, *D*, *E*, *G*, *H*, *M*, *N*, *O*), the difficulties of observation are less serious than in stouter

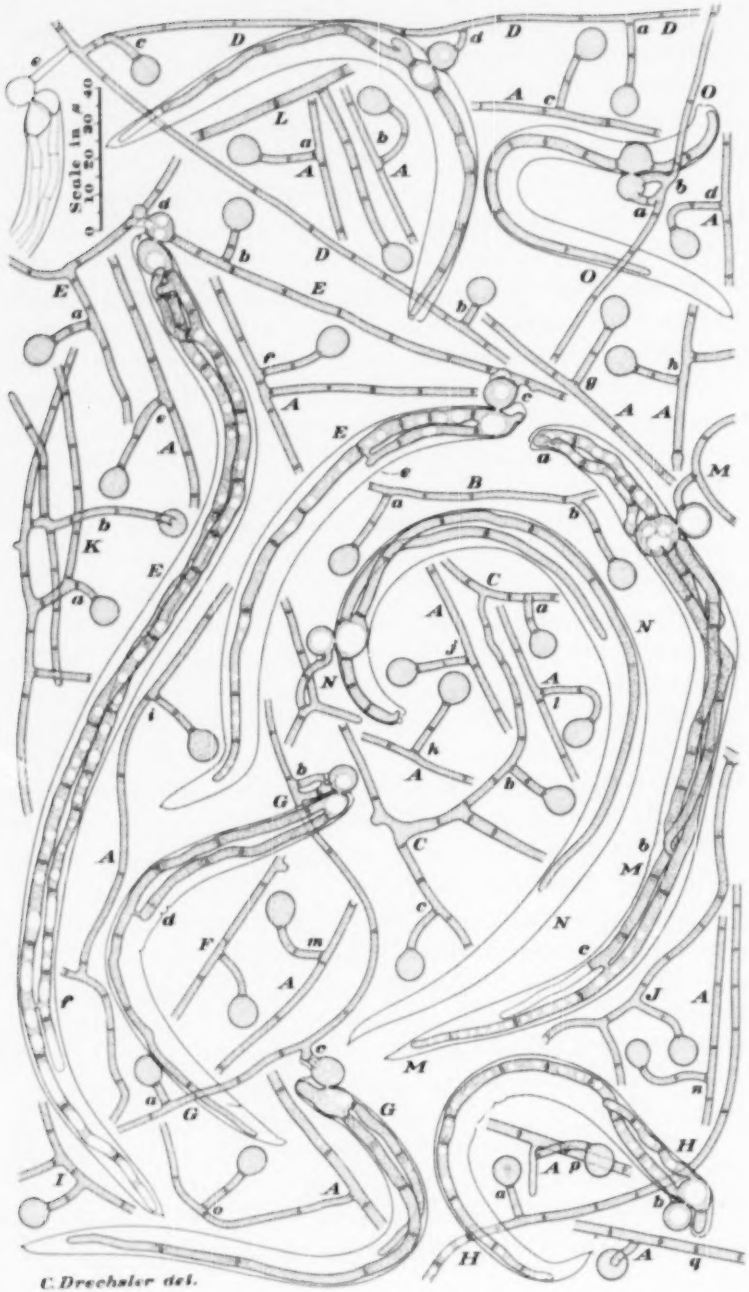

FIG. 13. *Dactylaria sclerohypha.*

animals. When the degenerating contents have in large part been absorbed by them, the assimilative hyphae, usually numbering from 1 to 3 in slender captives, become more clearly discernible, and reveal cross-walls at moderate intervals as well as scattered instances of vegetative fusion (FIG. 13, *E, e, f; G, d; M, a–c*). The hyphal segments, moreover, now frequently begin to show vacuoles. These vacuoles increase in volume as the animal's contents are further reduced (FIG. 13, *E, c, d; H; M*). Through continued withdrawal of protoplasm backward into the external mycelium, the assimilative hyphae are often completely evacuated of living substance, so that ultimately their empty membranous envelopes, together with the collapsing integument surrounding them and the empty membrane of the associated predacious organ (FIG. 13, *D, e*), gradually vanish from sight.

As in *Dactylaria haptotyla* many adhesive knobs that have been operative in capturing a nematode are found attached to the parent mycelial filament not only by the stalk whereon they were formed (FIG. 13, *O, a*) but also by a supplementary hyphal element (FIG. 13, *O, b*) of later origin. Sometimes a supplementary element intercalated between the knob and the parent hypha may become united laterally with the stalk (FIG. 13, *G, b*), which in its distal portion has been emptied of protoplasm presumably as the result of injury incurred from the struggles of the eelworm. Very frequently, however, where the supplementary element replaces a distal portion of the stalk, it is intercalated between the knob and the proximal living portion of the stalk (FIG. 13, *E, d; G, c; N*). Rather often, again, the stalk is found wholly emptied of protoplasmic material so that the supplementary hyphal element provides the only live communication between the knob and the mycelium (FIG. 13, *E, c*).

On nematode-infested materials, as also in pure culture on maizemeal agar, the fungus usually puts forth a scattered stand of conidiophores. The earliest conidiophores (FIG. 14, *A, a*) are sent up from procumbent mycelial hyphae, but when these have fallen over on the substratum (FIG. 14, *B, a; C; D, a*) after fulfilling their immediate function, they commonly give rise from a segment near the base to a secondary conidiophore (FIG. 14, *B, b; C; D, b*), which in turn may give rise to others of tertiary and

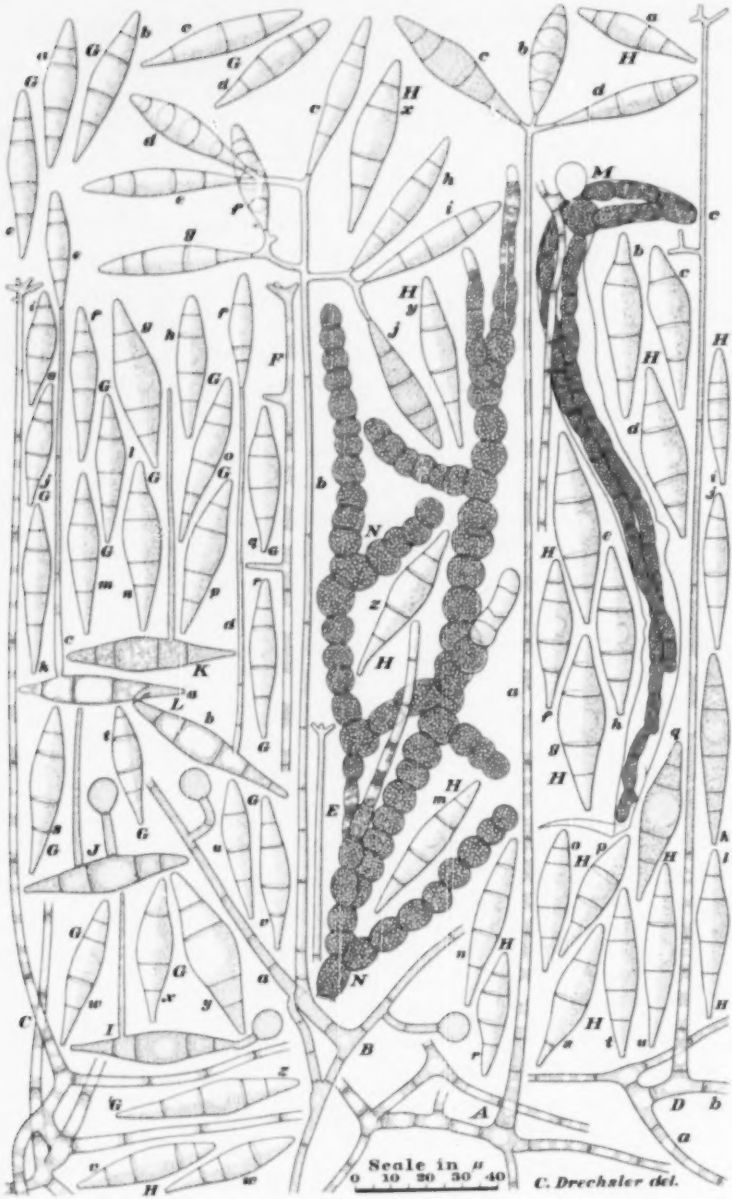

FIG. 14. *Dactylaria sclerohypha.*

higher orders (FIG. 14, *C; D, c*). As a rule the conidiophores are slender in proportion to their length, often seeming hardly sturdy enough to support the conidia, usually 3 to 8 in number (FIG. 14, *A, b–d; B, c–j*), which they hold aloft in loose capitate arrangement 200 to 350 μ above the substratum. In many instances their several sterigmatic spurs are all borne at the tip (FIG. 14, *C, E*), but not infrequently, again, one or more of the spurs are placed some distance below the tip (FIG. 14, *D, c; F*).

The conidia rather closely resemble those of *Dactylaria candida* and *Dactylaria haptotyla* in their dimensions and generally fusiform shape (FIG. 14, *G, a–z; H, a–z*). They are most commonly found divided by four cross-walls, although more than a few have three cross-walls (FIG. 14, *G, c, t, u, x; H, c, p, s, w*) and occasional specimens contain five (FIG. 14, *G, o; H, k*). Measurements of quadriseptate specimens have shown a range in length from 32 to 54 μ, and a range in width from 5.9 to 14.3 μ; and have given averages of 44.6 μ and 9.3 μ, respectively, for these two main dimensions. Measurements of the five component cells have yielded averages for lengths as follows: basal cell, 10.8 μ; parabasal cell, 8.4 μ; middle cell, 11.5 μ; penultimate cell, 6.2 μ; and apical cell, 7.7 μ. While in quadriseptate conidia the middle cell would thus seem generally somewhat longer as well as wider than the others, its length, on the whole, does not greatly exceed that of the basal cell, and in individual specimens is found appreciably smaller.

Under warm moist conditions conidia frequently put forth slender aerial filamentous outgrowths while they are still held aloft on the conidiophore. Similar outgrowths are often extended erectly after the spores have fallen on a moist substratum (FIG. 14, *I–K*). In many instances a filamentous outgrowth from a fallen conidium (FIG. 14, *L, a, b*) serves as a conidiophore (FIG. 14, *L, c, d*) in producing at its tip a secondary conidium (FIG. 14, *L, e, f*) usually of smaller dimensions than most conidia of primary origin but having similar shape and septation.

Fallen conidia that happen to lie close together (FIG. 14, *L, a, b*) very frequently become united through vegetative fusion manifestly of the same sort displayed abundantly in mycelial filaments and conidial apparatus throughout the series of clampless pre-

dacious hyphomycetes. Another vegetative attribute is shown by fallen conidia in their ready production of stalked adhesive knobs directly from one of their component cells (FIG. 14, *I, J*). Development of like predacious organs on detached conidia was noted earlier in the descriptive account of *Dactylella asthenopaga*. As might well be expected, stalked adhesive knobs are also produced occasionally on conidiophores (FIG. 14, *B, a*) that have fallen over on moist substratum while they were still largely filled with living protoplasm.

After the fungus has been producing conidia in nematode-infested cultures for a period of 10 to 15 days, it often diverts much of its substance toward the formation of chlamydospores or indurated portions of mycelium. Once the change in reproductive tendency has set in, the assimilative hyphae in many instances will not convey their protoplasm to the external mycelium, but instead will retain it within their constituent segments to be thickly interspersed with globulose reserve materials elaborated from the digestible contents of the eelworm (FIG. 14, *M*). In taking on a durable state the hyphal segments here usually become noticeably but not markedly widened. The infective cell, which likewise undergoes induration, reveals generally no further increase in size.

Development of resting bodies through transformation of assimilative hyphae within captured animals has so far never been observed in any other clampless nematode-capturing hyphomycete, nor, for that matter, in the four members of the Zoöpagaceae known to capture eelworms (**10, 11, 14, 17**), nor, again, in the two species of similar biological habit that have been described in the clamp-bearing genus *Nematoctonus* (**18, 20**). Among the various hyphomycetes attacking eelworms after the usual manner of parasites, by means of hyphae arising through germination of affixed or ingested spores, analogous internal development of resting bodies has become familiar to me only in the production of chlamydospores by the ubiquitous *Harposporium anguillulae* Lohde. In the present fungus, besides, scattered filaments of the external mycelium, together with many of their branches, undergo induration to form often rather extensively ramified sclerotioid bodies (FIG. 14, *N*). The hyphal segments here, both in the main filaments and in the branches, frequently become distended to three times their original

width, so that the indurated cells are generally of subspherical shape, and in many instances may even measure more in transverse diameter than in length. As in the similar sclerotioid bodies of *Dactylella heterospora*, the more strongly distended cells, which here may form chains over 1 mm. long, are always densely filled with globulose contents, while the cells that have become only slightly distended show correspondingly less internal modification.

A term having reference to the filamentous character of its resting bodies may serve appropriately as specific epithet for the fungus, especially as the resting bodies often occur in a somewhat web-like arrangement.

Dactylaria sclerohypha sp. nov.

Mycelium sparsum; hyphis incoloratis, mediocriter septatis, plerumque 1.5–3.2 μ crassis, hic illic ex ramulo recto vel curvato, 5–50 μ longo, 2–2.5 μ crasso, in 1–4 (saepissime in 1 vel 2) cellulis consistente, bullas tenaces globosas vel ellipsoideas 8.3–10 μ longas, 7.2–8.6 μ crassas, singillatim emittentibus; his bullis ad vermiculos nematoideos inhaerentibus, ita animalia tenentibus, integumentum eorum perforantibus, tuber mortiferum intrudentibus, hyphas plerumque 2–4.5 μ crassas intus evolventibus quae carnem exhauriunt. Hyphae fertiles incoloratae, erectae, saepius 5–10 septatae, 200–350 μ altae, basi 3.3–5 μ crassae, sursum leniter attenuatae, apice 1.4–2 μ crassae, primum saepe simplices et in unum conidium abeuntes, postea apice vulgo aliquid ramosae denique 2–8 conidia in capitulum laxum ferentes; conidiis incoloratis, vulgo fusoideis, apice anguste rotundatis, basi truncatis, plerumque 32–54 μ (saepius circa 44.6 μ) longis, 5.9–14.3 μ (saepius circa 9.3 μ) crassis, 3–5 septatis, saepissime quadriseptatis denique cellula antepenultima eorum plerumque crassiore et longiore quam aliis cellulis, post disjunctionem bullam tenacem vel hypham fertilem interdum emittentibus; hyphis fertilibus germinationis simplicibus saepius circa 100 μ altis, basi 2–2.5 μ crassis, sursum leniter attenuatis, apice circa 1.2 μ crassis, saepius biseptatis vel triseptatis, unum conidium ferentibus; conidiis ordinis secundi fusiformibus plerumque triseptatis vel quadriseptatis, vulgo circa 32 μ longis et 6.5–7 μ crassis. Corpora perdurantia intra animalis atque in materia circumdanti orta, filiformia, vulgo ramosa, paene incolorata vel flavidula, protoplasmatis valde guttulosi repleta, saepe 0.2–1 mm. longa, in cellulis plerumque 8–15 μ longis et 3–13 μ crassis consistentia.

Vermiculos nematoideos multarum specierum capiens consumensque habitat in materiis plantarum putrescentibus prope Roanoke, Virginia.

Mycelium scanty; vegetative hyphae colorless, septate at moderate intervals, mostly 1.5 to 3.2 μ wide, often especially in the presence of nematodes giving rise here and there on straight or curved stalks, 5 to 50 μ long and 2 to 2.5 μ wide, usually uniseptate or biseptate but sometimes triseptate or quadriseptate, to solitary ad-

hesive knobs, globose or ellipsoidal in shape, commonly 8.3 to 10 μ long and 7.2 to 8.6 μ wide; these knobs holding fast to nematodes, thus capturing the animals, then perforating the integument of each captive and intruding a globose infective body from which assimilative hyphae, mostly 2 to 4.5 μ wide, are extended lengthwise to appropriate the fleshy contents. Conidiophores colorless, erect, often containing 5 to 10 cross-walls, frequently 200 to 350 μ high, 3.3 to 5 μ wide at the base, gradually tapering upward to a width of 1.4 to 2 μ at the tip, at first often simple and terminating in a single conidium but later often provided distally with short branches or spurs and then bearing 2 to 8 conidia in loose capitate arrangement; conidia colorless, commonly spindle-shaped, narrowly rounded at the tip, truncate at the base, mostly 32 to 54 μ (average about 44.6 μ) long, 5.9 to 14.3 μ (average about 9.3 μ) wide, containing from 3 to 5 cross-walls but most often divided by 4 septa into 5 cells whereof the middle one is widest and usually longest, after being abjointed sometimes putting forth a stalked adhesive knob or a conidiophore; conidiophores of germinative origin usually simple, often biseptate or triseptate, about 100 μ high, 2 to 2.5 μ wide at the base, tapering gradually upward to a width of 1.2 μ at the tip, whereon is borne a single secondary conidium; secondary conidia spindle-shaped, mostly triseptate or quadriseptate, commonly about 32 μ long and 6.5 to 7 μ wide. Resting bodies formed within the animal's integument and also externally, filamentous, frequently branched, nearly colorless or faintly yellowish, often 0.2 to 1 mm. long, composed of cells mostly 8 to 15 μ long and 3 to 13 μ wide that are filled with densely globuliferous protoplasm.

Capturing and consuming nematodes of different species it occurs in decaying plant materials near Roanoke, Virginia.

A CLAMPLESS HYPHOMCETE CAPTURING NEMATODES IN THICK NON-CONSTRICTING RINGS

The several maizemeal-agar plate cultures which after being planted with leaf mold from woods near Greensboro, North Carolina, permitted abundant development of *Dactylella aphrobrocha*, were occupied rather extensively also by a number of other clampless hyphomycetes of like predacious habit. Among these allied fungi was a species that produced three-celled non-constricting rings (FIG. 3, *S*; *T*; *U*, *a*; *V*) and globose or ellipsoidal unicellular knobs (FIG. 3, *U*, *b*) on delicate, mostly bicellular stalks often about 25 μ in length. While the development of these two types of pre-

dacious organs on the same mycelium offered general parallelism with development in *Dactylella lysipaga*, *Dactylella leptospora*, and *Dactylaria candida*, the rings here could immediately be recognized as being larger and more massive than those of the three species mentioned. They measured from 20 to 26 μ in outside diameter and thus with respect to this dimension exceeded the rings of the three similar species by approximately 5 μ. In respect to thickness of the three cells composing them they ranged from 3 to 5 μ, their more usual transverse measurement of approximately 4 μ exceeding the corresponding measurement in the three known similar forms by about 1 μ. On the whole they would appear, indeed, very nearly equal in size and volume to the constricting rings of *Dactylella stenobrocha*. This approximate equality provides an exception to the marked dimensional disparity generally observable among annulated members of the predacious series, between the rather frail non-constricting rings and the conspicuously sturdy constricting rings.

For the most part the rings operated like those of *Dactylella lysipaga*, *Dactylella leptospora*, and *Dactylaria candida*. In some instances the slender supporting stalk withstood the frantic struggles of the ensnared nematode, though often incurring injury distally (FIG. 3, *W*, *a*) that needed to be repaired through intercalation of a new hyphal connection (FIG. 3, *W*, *b*). In other instances the stalk broke, permitting the animals, now tightly encircled by the ring (FIG. 3, *X*, *a*) to continue moving about for a few hours longer. In either event the ring would soon put forth from its inner surface a narrow protuberance which after penetrating the animal's integument gave rise within the fleshy interior to a globose infective body. Owing apparently to the greater volume of protoplasm contained in the ring the infective body here continued to enlarge, as a rule, until it occupied the whole width of the eelworm, whereas in the three species with smaller rings it commonly extends only partly across the animal. The virtually complete severance of musculature and organs promptly resulted in disablement of the eelworm, thereby making possible early extension of assimilative hyphae from the infective body throughout the fleshy interior. Where a nematode, after breaking the stalk, moved some distance from the predacious mycelium before succumbing to infection, its

substance was utilized by the fungus to put forth from the rings one or more external hyphae that grew out to establish a new tract of predacious mycelium (FIG. 3, *X*, *b*).

The knobs were never observed taking any part in the destruction of eelworms. It may be presumed that in this species as also in *Dactylella lysipaga*, *Dactylaria haptospora*, and *Dactylaria candida*, these relatively small organs are designed to function under conditions somewhat different from those suitable for the operation of non-constricting rings—under conditions apparently not often occurring in agar plate cultures of the sort employed by me. Although the mycelium bearing the two kinds of predacious organs was kept under observation until it degenerated from age, it has not so far been found giving rise to conidiophores and conidia, nor has it been seen associated with any other type of reproductive apparatus. The fungus would appear distinct from *Anulosporium nematogenum* Sherbakoff (**30**) in view of the larger dimensions of its rings. Its further characterization is deferred until the discovery of a conidial stage may permit reference to the proper genus in the predacious series of hyphomycetes.

GONATOBOTRYS SIMPLEX AND ITS RELATIONSHIP TO THE PREDACIOUS HYPHOMYCETES

In the classic "Pracht-Flora" wherein Corda (**9**) more than a century ago published the original account of his *Arthrobotrys superba* he likewise presented the original description of his *Gonatobotrys simplex*. The two fungi were set forth as being similar in their development of conidiophores with swollen nodes, in the spiral arrangement of warty sporiferous protuberances on the nodes, and in their production of oblong spores in successive capitate clusters. Owing, however, to difference in cellular make-up of their spores each was presented, in conformity with the requirements of taxonomic practice, as the type of a separate new mucedinous genus; unseptate conidia being thereby specified for the genus *Gonatobotrys*, and uniseptate conidia for *Arthrobotrys*. De Bary made the two genera more familiar by citing them together, in both editions of his well known manual on fungi (**1**: 46, 47; **2**: 50), as exemplifying successive development of conidial

clusters on a repeatedly prolonged conidiophore. The unseptate conidia specified for it gave occasion for assignment to *Gonatobotrys* of such diverse species as *G. microspora* Rivolta (**28**) and *G. pallidula* Bresadola (**5**): the former, with its continuous mycelium, having subsequently been recognized as a phycomycete and enrolled among the Mucorales under the binomial *Cunninghamella microspora* (Riv.) Matruchot (**26**: 56; **31**: 168); whereas the latter, with clamp-connections in its mycelial filaments, has been transferred to the resupinate basidiomycetes as *Peniophora pallidula* (Bres.) Bres. apud Bourdot et Galzan (**4, 29**). Neither the unhappy application of *Gonatobotrys*, nor the persistent confusion of *Arthrobotrys oligospora* with *Trichothecium roseum* Link to which some discussion was given earlier (**12**: 469–472), can be held to have upset the parallelism pointed out by Corda, as far as this parallelism concerns forms wherein the sporiferous and mycelial hyphae are divided by ordinary cross-walls. The possibility is not to be dismissed that among septate clampless hyphomycetes such parallelism might in some instances, if not generally, derive from close kinship, and thus might be associated with biological similarities. For this reason, after *A. superba*, together not only with *A. oligospora* and other congeners but also with many related hyphomycetes producing multiseptate spores of various sizes and shapes, was found subsisting habitually through capture of nematodes, cultures infested with eelworms have been observed closely for the development of predacious fungi referable to *Gonatobotrys*. In the course of more than a decade, however, no clampless hyphomycete bearing unicellular conidia in successive or in solitary clusters has ever been found preying on eelworms in infested agar cultures planted with decaying vegetable materials of different kinds. Therefore, when some years ago a true *Gonatobotrys* appeared abundantly on tomato (*Lycopersicon esculentum* Mill.) and muskmelon (*Cucumis melo* L.) leaves in the laboratory, opportunity was taken to try it out on nematodes, and also to grow it in pure culture for comparison with familiar members of the predacious series of clampless hyphomycetes.

The muskmelon leaves here in question were gathered in a field near Baltimore, Maryland, on July 13, 1942, and bore large dry lesions due evidently to *Alternaria cucumerina* (Ell. & Ev.) J. A.

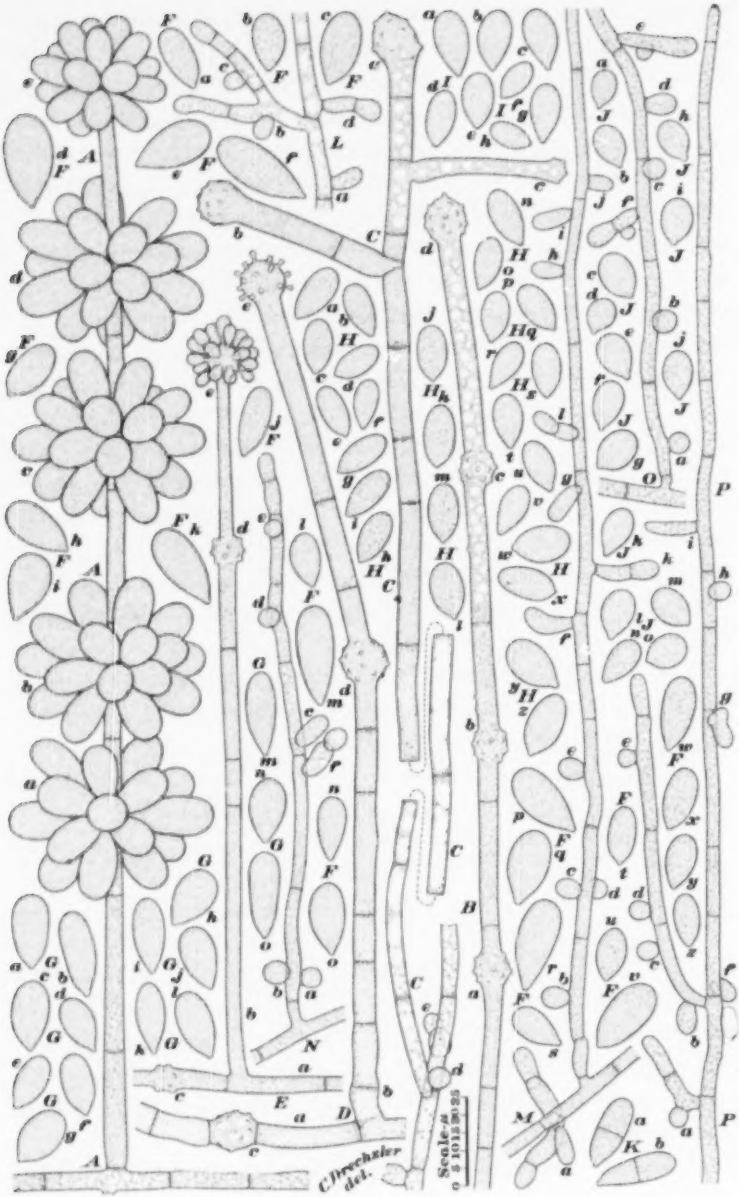

FIG. 15. *Gonatobotrys simplex.*

Elliott. Rather similar lesions attributable to the early-blight fungus, *Alternaria Solani* (Ell. & Martin) Sor., were present on the tomato leaves when these were collected in a field near Beltsville, Maryland, on July 25, 1942. Soon after their removal from the plants the leaves were arranged in a moist chamber and stored at a temperature of 18° C. On examining them 10 days later they were found covered extensively with several different molds. The *Gonatobotrys* appeared as a white compact layer covering irregular patches mostly 1 to 5 square centimeters in area. It offered to the naked eye a delicately granular appearance; the individual granules becoming recognizable under the microscope as conidial clusters (FIG. 15, *A*, *a–c*) borne at intervals of 20 to 100 μ on erect or drooping conidiophores mostly 4.5 to 7 μ wide. When small tracts of the sporulating layer were mounted in water under a cover-glass, virtually all mature conidia became abjointed from the conidiophores, exposing to view the denuded nodes, usually 8 to 12 μ wide, and beset with truncated wart-like protuberances (FIG. 15, *B*, *a–d*). In pure culture on maizemeal agar the conidiophores frequently bore at some little distance below the expanded sporiferous tip (FIG. 15, *C*, *a*) one or more lateral branches each of which similarly terminated in a fertile tip often perceptibly swollen (FIG. 15, *C*, *b*, *c*). Such branching, as a rule, was associated with less abundant development of spore clusters on the repeatedly prolonged axial hypha. Older conidiophores, both in pure culture (FIG. 15, *D*, *a*) and on the softened leaves (FIG. 15, *E*, *a*), often declined to the substratum, and would then send up an erect branch (FIG. 15, *D*, *b*; *E*, *b*) in a position usually a little removed from any of its nodes (FIG. 15, *D*, *c*; *E*, *c*). Through the production of spore clusters at successive nodes (FIG. 15, *D*, *d*, *e*; *E*, *d*, *e*) these branches served as secondary conidiophores.

The development of the several conidia in a cluster appears here more strictly simultaneous than in any of the species of *Arthrobotrys* and *Dactylaria* that are known to capture eelworms. At the earliest stage in the formation of a cluster the distended tip of the conidiophore is found bearing from twenty to thirty protuberances, all equally minute (FIG. 15, *D*, *e*); the appearance then given recalling young sporophores of *Cunninghamella echinulata* (Matr.) Thaxter. Growth of the protuberances proceeds steadily,

each of them keeping pace accurately with the enlargement of its fellows (FIG. 15, *A, e; E, e*), so that all reach definitive size and are ready to be abjointed as conidia at the same time (FIG. 15, *A, a–d*). Conidia produced on tomato and muskmelon leaves are generally of an elongated obovoid shape though noticeably apiculate at the basal end (FIG. 15, *F, a–z; G, a–o; H, a–z; I, a–h*). They measure commonly from 12 to 29 μ in length and from 7 to 13.5 μ in greatest width. Conidia produced in pure culture on maizemeal agar are of more broadly obovoid conformation and of smaller dimensions, their length varying ordinarily from 11 to 15 μ, and their greatest width from 7 to 10 μ (FIG. 15, *J, a–o*). The unseptate condition is always characteristic of the conidia, regardless of the substratum on which they are formed. However, in some mounts of material produced on tomato leaves a few uniseptate specimens (FIG. 15, *K, a, b*) were found among many thousands wherein a cross-wall was lacking. These closely resembled the conidia of *Arthrobotrys superba* and may, perhaps, have been present as an incidental admixture of that species.

In pure culture on maizemeal agar the fungus grows more slowly than the larger number of clampless hyphomycetes that are known to subsist by capturing eelworms. Nevertheless in 10 to 15 days it usually gives rise to aerial hyphae and conidiophores in easily recognizable quantity. To the naked eye the aerial weft offers a fairly pronounced orange coloration decidedly deeper in shade than the pink coloration characteristic of the conidial apparatus formed so abundantly in pure cultures of *Dactylaria polycephala*. When sizable portions of agar permeated with young mycelium of the fungus were transferred to agar plate cultures infested with various eelworms, amoebae, and testaceous rhizopods, no predacious or parasitic characteristics came to light. From such trials, however, it might be premature to deny all possibility of the fungus subsisting on animals, since in pure culture its mycelial hyphae are regularly found bearing globose or digitate lateral branches that consist commonly of one (FIG. 15, *L, a–c; M, a–j; N, a–e; O, a–e; P, a–i*) or two (FIG. 15, *L, d; M, k, l; N, f; O, f*) cells, and thereby in some degree resemble the adhesive outgrowths of *Dactylella cionopaga*. Though these curious lateral branches have shown no efficacy for the destruction of the few types of animals present in my cultures,

the large variety of minute animals capable of multiplying on the lower leaves of vegetable crop plants, especially during prolonged periods of wet weather, might well offer scope for many biological relationships not hitherto revealed to sight.

As my fungus when developing on leaves in the dark at a temperature of 18° C. agrees rather well with Corda's description of *Gonatobotrys simplex*, it is assigned to that ancient species,—the assignment appearing further justified through observations by Coemans (**7**), Harz (**23**), and Matruchot (**25**) which show that wide variability with respect to coloration and branching is usual here. On grounds of priority Corda's binomial seems preferable to some others with which it has been held synonymous; for *Gonatobotrys* clearly antedates the genus *Oedocephalum* Preuss (**27**: 131) as well as the genus *Desmotrichum* Léveillé (**24**), while similarly the species *G. simplex* antedates *G. flava* Bon. (**3**: 105), *G. ramosa* Riess ex Fresenius (**21**), and *Oedocephalum roseum* Cooke (**8**). The relationship of the fungus to *Arthrobotrys* and to the series of clampless predacious hyphomycetes remains problematical. Although the general parallelism in manner of conidial development argues for kinship, the protoplasm in the conidiophore seems of markedly different texture from that in most mucedinous forms known to capture eelworms. Woronin bodies are either absent here or far less readily discernible than in the familiar species of *Arthrobotrys*. The rather distinctive odor given off by maizemeal-agar cultures of all known nematode-capturing species of *Arthrobotrys* has never been detected in any cultures of the *Gonatobotrys*.

Since Corda set forth *Gonatobotrys simplex* as growing parsitically on *Helmisporium tenuissimum* Nees, it was deemed appropriate to try out my fungus on two dematiaceous forms—*Alternaria Solani* and *Alternaria tenuis* Nees—that developed abundantly nearby on the same tomato leaves. Accordingly sizable portions of agar well permeated with young mycelium were taken from pure cultures of the *Gonatobotrys* and placed on maizemeal-agar plate cultures of the two species of *Alternaria*. In these trials the *Gonatobotrys* showed no capacity for parasitizing the two dematiaceous forms.

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EXPLANATION OF FIGURES

FIG. 1. *Dactylella stenobrocha* as found developing in nematode-infested agar plate cultures; drawn to a uniform magnification with the aid of a camera lucida; × 500 throughout. *A*, Portion of mycelial hypha bearing an open predacious ring in its original position. *B–E*, Portions of hypha, each bearing an open predacious ring that is turned sideways, thereby showing advantageously its three arcuate cells and the two cells making up the stalk. *F*, Longer portion of hypha bearing two open predacious rings, *a* and *b*, that are turned sideways, thereby revealing their cellular make-up advantageously. *G*, Portion of hypha bearing a predacious ring that has closed emptily without capturing an eelworm; the ring being turned sideways shows advantageously the shape and vacuolate condition of the contracted arcuate cells. *H*, Portion of hypha with a ring that has contracted in capturing a specimen of *Plectus* sp.; two stout protuberances, one extending from the closed ring forward within the animal, and the other extending from the closed ring backward within the animal, represent assimilative hyphae in an early stage of development. *I*, Portion of hypha with a ring that has contracted in capturing a specimen of *Plectus* sp.; the two assimilative hyphae, one extending forward and the other backward within the animal, have attained their definitive length, with each showing some swelling of its terminal segment. *J*, Portion of hypha bearing a closed ring that holds a captured specimen of *Plectus* sp.; the two long assimilative hyphae extending to the animal's head and tail, respectively, each show some enlargement of the two terminal segments and the pronounced vacuolization of all segments that comes with advanced exhaustion of fleshy substance. *K*, Portion of prostrate hypha, *a*, with two erect conidiophores, *b* and *c*, whereon are borne the conidia *d* and *e*, respectively; each conidiophore being shown, from want of space, in three parts whose proper connection is indicated by broken lines. *L*, Portion of prostrate hypha, *a*, with a fallen conidiophore, *b*, that has given rise to an

erect secondary conidiophore, *c*, which bears at its tip the well developed conidium *d*; the conidiophore *c* being shown, from lack of space, in three parts whose proper connection is indicated by broken lines. *M*, Short basal portion of a fallen conidiophore, *a*, from which has been sent up the erect secondary conidiophore *b*, bearing terminally a conidium, *c*; the conidiophore *b* being shown, from lack of space, in three parts whose proper connection is indicated by broken lines. *N*, Mature detached conidia showing usual variations in size and shape, together with variations in number and position of septa as follows: *a–c*, single septum near the proximal end; *d–o*, two septa near the proximal end; *p–s*, two septa symmetrically placed, one near the proximal end, the other near the distal end; *t–z*, three septa, two near the proximal end, one near the distal end. *O*, Two conidia, *a* and *b*, each with three septa placed near the proximal end.

FIG. 2. *Dactylella stenobrocha* as found developing in nematode-infested agar plate cultures; drawn to a uniform magnification with the aid of a camera lucida; × 500 throughout. *A*, Portion of mycelial hypha bearing two predacious rings, *a* and *b*, in their original positions. *B–E*, Portions of hypha, each bearing a predacious ring that is turned sideways, thereby showing its cellular make-up advantageously. *F*, Short portion of hypha with a closed predacious ring nearly in its original position. *G*, Portion of hypha with a predacious ring that has closed in capturing a specimen of *Plectus* sp.; no penetration into the animal has yet taken place. *H*, Portion of hypha whereon is borne a predacious ring, *a*, that closed in capturing a specimen of *Plectus* sp. and then invaded the animal with three assimilative hyphae, two of which are rather markedly distended in their two terminal segments; nearby is attached a second predacious ring, *b*, that has closed emptily without capturing any eelworm. *I*, Portion of hypha with a predacious ring that has closed in capturing a specimen of *Plectus* sp., and has extended an assimilative hypha forward, and another one backward through the animal's body; the last two segments of the hypha directed forward each showing marked enlargement. *J*, Portion of hypha with a predacious ring that has closed in capturing a specimen of *Plectus* sp., and has given rise to assimilative hyphae extending forward and backward through the animal's body; *a*, anastomosis of two assimilative hyphae. *K*, *L*, Portions of prostrate hypha, each with an erect conidiophore, *a*, near the tip of which is shown a conidium, *b*, that was abjointed from it; the conidiophore *a* in each instance is shown, from lack of space, in three parts whose proper connection is indicated by broken lines. *M*, Mature detached conidia, showing usual variations in size and shape, together with variations in number and position of septa as follows: *a*, single septum near basal end; *b–m*, two septa near the basal end; *n*, *o*, two septa symmetrically placed, one near the proximal end, the other near the distal end; *p–z*, three septa, two of them near the proximal end, the other near the distal end. *N*, Conidium with three septa, two of them near the proximal end, the other one near the distal end. *O*, Detached triseptate conidium that has sent up an aerial hypha from its small apical segment. *P*, Detached biseptate conidium that has sent up an aerial hypha from its basal segment. *Q*, Abjointed triseptate conidium that has sent up two aerial hyphae, one from its small parabasal segment, the other from its small apical segment. *R*, De-

tached conidium that has put forth a germ tube from a position next to its base.

FIG. 3. Drawn to a uniform magnification with the aid of a camera lucida; × 500 throughout. *A, B,* Portions of hyphae, each with a denuded conidiophore of *Dactylella stenobrocha* as found in nematode-infested agar plate cultures; each conidiophore being shown, from a lack of space, in three parts whose proper connection is indicated by broken lines. *C–H, Dactylella stenobrocha* as found developing in pure culture on maizemeal agar: *C–E,* Portions of hyphae, each with a conidiophore from whose tip a single conidium has been abjointed. *F,* Uniseptate conidia, *a–o,* showing usual variations in size and shape. *G,* Biseptate conidia showing usual variations in size and shape, some of them (*a–h*) having both cross-walls placed near the proximal end, and others (*i–p*) having one cross-wall near each end. *H,* Triseptate conidium. *I,* Detached conidium of *Dactylella aphrobrocha* produced in a nematode-infested agar plate culture that had been planted with deciduous leaf mold gathered near Fairfax, Virginia, in November, 1942. *J–R, Dactylella cionopaga* as found developing in nematode-infested agar plate cultures that had been planted with deciduous leaf mold gathered near Butternut, Wisconsin, in September, 1938: *J,* Portion of hypha bearing a simple three-celled predacious outgrowth, *a,* and a branched seven-celled predacious outgrowth, *b*. *K,* Portion of hypha with three predacious outgrowths, *a–c,* to which is affixed a small stylet-bearing nematode; a globose infective body intruded from the two-celled outgrowth *c* has given rise within the animal to a growing assimilative hypha. *L,* Portion of hypha with four predacious outgrowths, *a–d,* of which three (*b–d*) have jointly captured a small nematode (probably referable to *Plectus* sp.) and have each intruded a bulbous infective body into the captive; the several assimilative hyphae extended from the infective bodies have become joined through vegetative fusion. *M, N,* Detached triseptate conidia. *O–Q,* Detached quadriseptate conidia. *R,* Quadriseptate conidium germinating by two polar germ tubes. *S–X,* Unnamed hyphomycete found capturing nematodes in agar plate cultures that had been planted with leaf mold gathered near Greensboro, North Carolina, in December, 1947. *S, T,* Portions of mycelial hyphae, each bearing a non-constricting ring. *U,* Portion of mycelium bearing a non-constricting ring, *a,* and a globose adhesive cell, *b*. *V,* Portion of hypha with a non-constricting ring. *W,* Portion of hypha with a non-constricting ring that after having operated in capturing a nematode (possibly referable to *Wilsonema* sp.) has intruded into the animal a globose infective body from which four assimilative hyphae are being extended lengthwise through the fleshy interior; the narrow stalk, *a,* originally supporting the ring has been evacuated distally, and a new connection, *b,* has been formed. *X,* Small eelworm that after being captured in a non-constricting ring, *a,* tore loose the encircling organ but was nevertheless later killed through intrusion of a globose infective body from which it was then invaded throughout by assimilative hyphae; a mycelial hypha, *b,* grew out externally from the ring.

FIG. 4. *Dactylella aphrobrocha* as found developing in nematode-infested agar plate cultures; drawn to a uniform magnification with the aid of a camera lucida; × 500 throughout. *A,* Portion of hypha bearing a large predacious ring in its original position. *B,* Portion of hypha bearing an open

predacious ring, *a*, in its original position, and another such ring, *b*, displaced sideways a little; the stalk supporting ring *b* is unusual in length, and in being composed of three cells. *C*, Portion of hypha with two open predacious rings, *a* and *b*, both being turned sideways out of their original positions. *D*, Portion of hypha bearing an open predacious ring of relatively small size. *E*, Portion of hypha with a rather large open predacious ring that is turned sideways a little. *F–H*, Portions of hypha, each bearing an open predacious ring that is turned flatwise, thereby advantageously showing its cellular make-up. *I*, Portion of hypha with two open predacious rings, *a* and *b*, both being turned flatwise. *J*, Portion of hypha with a closed predacious ring that is turned flatwise. *K*, Portion of hypha with a predacious ring that has closed in capturing an eelworm (possibly referable to *Plectus parvus* Bastian) and has given rise to assimilative hyphae within the animal. *L*, Portion of hypha with a predacious ring that after capturing an eelworm has extended several assimilative hyphae lengthwise through its body; these hyphae have very nearly exhausted the animal's fleshy substance, and are becoming vacuolate from transfer of protoplasm to the external mycelium. *M*, *N*, Portions of prostrate hyphae, each bearing an erect conidiophore, *a*, near the tip of which is shown a conidium, *b*, that was abjointed from it; the conidiophore *a* being shown in each instance, from lack of space, in three parts whose proper connection is indicated by broken lines. *O*, Random assortment of detached mature conidia, showing usual variations in size and shape, together with variations with respect to number and position of septa as follows: *a–c*, three septa present, two of them placed near the proximal end, and one near the tip; *d*, three septa present, one of them placed near the proximal end, and two near the tip; *e–z*, four septa present, in symmetrical arrangement, two being placed near each end. *P*, *Q*, Symmetrically quadriseptate conidia, each of which has extended an aerial hypha from its rather small parabasal cell. *R*, Symmetrically quadriseptate conidium that has put forth two aerial hyphae, one from the penultimate segment, the other from the parabasal segment. *S*, *T*, Symmetrically quadriseptate conidia, each germinating by putting forth a germ tube from its basal and its apical cell.

FIG. 5. *Dactylella aphrobrocha* as found developing in nematode-infested agar plate cultures; drawn to a uniform magnification with the aid of a camera lucida; × 500 throughout. *A*, Portion of mycelial hypha bearing a large open predacious ring in its original position. *B*, Portion of hypha with an open predacious ring that has been turned sideways to a considerable degree. *C–H*, Portions of hyphae, each with an open predacious ring that is turned flatwise, thereby showing advantageously usual variations in size and shape of the arcuate cells, the stalk cells, and the aperture. *I*, Several successive stages in the formation of a predacious ring; *a*, simple branch of circinate curvature, with two cross-walls delimiting the two cells of the stalk; *b*, same branch 1½ hours later, showing its tip directed toward the tip of a secondary spur that is growing out from the distal segment of the stalk; *c*, same branch 30 minutes later than *b*, showing its tip in contact with the tip of the spur, and presence of a third cross-wall, which delimits the proximal arcuate cell; *d*, same branch 30 minutes later than *c*, showing fusion of tip and spur, and continued homogeneous appearance of protoplasmic contents throughout the ring and its stalk; *e*, same branch 17 hours later than *d*,

showing completed ring in functional condition, each of the arcuate cells containing an irregular elongated lacuna of obscurely globulose or foamy composition. *J*, Portion of hypha with a rather small predacious ring in lateral view; the stalk here being exceptional in consisting of three cells. *K*, Portion of hypha with a predacious ring which after capturing a small eelworm (possibly referable to *Plectus parvus*) has given rise to young assimilative hyphae within the animal. *L*, Portion of hypha with a predacious ring which after capturing a small eelworm has given rise to assimilative hyphae extending lengthwise through the animal. *M*, Portion of mycelium with a predacious ring which after capturing an eelworm of larger size has given rise to assimilative hyphae extending lengthwise through the animal; the stalk *a* originally supporting the ring having suffered injury, a new connection, *b*, has been formed between the mycelial hypha and one of the swollen cells. *N*, Portion of hypha with a predacious ring which after capturing an eelworm gave rise to assimilative hyphae extending lengthwise through its body; the original stalk, *a*, and two of the swollen cells having incurred destructive injury, a new hyphal connection, *b*, has been formed between the parent mycelial filament and the irregular proximal infective cell of the assimilative hyphal system. *O*, Portion of prostrate hypha bearing an erect conidiophore, *a*, which on its axial tip and on the tip of a branch, *b*, bore the conidia *c* and *d*, respectively; the conidiophore, from lack of space, being shown in three parts whose proper connections are indicated by broken lines. *P*, Random assortment of conidia, showing usual variations in size and shape, together with variations in number and position of septa as follows: *a–c*, three septa present, whereof two are placed near the proximal end, and one near the distal end; *d–w*, four septa present in symmetrical arrangement, two being placed near each end.

FIG. 6. *Dactylella cionopaga* drawn to a uniform magnification with the aid of a camera lucida; × 500 throughout. *A*, *B*, Portions of hypha, each bearing a unicellular dome-shaped adhesive outgrowth. *C*, Portion of prostrate hypha bearing a unicellular columnar adhesive outgrowth, *a*, two bicellular adhesive outgrowths, *b* and *c*, and a five-celled adhesive arch, *d*. *D*, Portion of prostrate hypha bearing a four-celled columnar adhesive outgrowth, *a*, together with a three-celled adhesive arch, *b*, and a six-celled adhesive arch, *c*; the adhesive arch *b* is surmounted by a two-celled adhesive process. *E*, Portion of prostrate hypha with a low adhesive protuberance, *a*, arising from one of its segments; on the hypha are borne further a two-celled adhesive outgrowth, *b*, three four-celled columnar adhesive outgrowths, *c–e*, and an eight-celled network of two meshes, *f*. *F*, Portion of prostrate hypha with a six-celled adhesive outgrowth. *G*, Portion of hypha with a three-celled adhesive outgrowth which after capturing a slender eelworm (possibly referable to *Plectus* sp.) has intruded three bulbous excrescences into the animal; the excrescence intruded from the basal segment having given rise to a short, growing assimilative hypha. *H*, Portion of prostrate hypha with a three-celled adhesive process that after capturing a slender eelworm has intruded into the animal a bulbous excrescence from which a short, growing assimilative hypha has been extended. *I*, Portion of prostrate hypha bearing a two-celled columnar adhesive outgrowth that after capturing a slender eelworm has intruded into the animal a bulbous excrescence from which one

assimilative hypha has been extended forward and another backward through the fleshy body. *J*, Portion of prostrate hypha with two bicellular adhesive outgrowths, *a* and *b*, each of which has intruded a bulbous excrescence into a slender eelworm that they had captured jointly; assimilative hyphae have begun growing out from each of the intruded bodies. *K*, Portion of prostrate hypha with a unicellular adhesive outgrowth, *a*, and a two-celled adhesive outgrowth, *b*, which have each intruded a bulbous excrescence into a slender eelworm captured jointly by them; assimilative hyphae extended from the two bulbous bodies have become united by an anastomosing connection, *c*. *L–N*, Portions of prostrate hyphae, each bearing an erect conidiophore from which a conidium has been abjointed. *O*, Mature detached conidia, showing usual variations in size and shape, together with variations with respect to septation as follows: *a*, two cross-walls present; *b–f*, three cross-walls present; *g–n*, four cross-walls present; *o*, *p*, five cross-walls present; *q*, six cross-walls present. *P*, Fallen conidium that has put forth a two-celled adhesive outgrowth, *a*, from its distal end. *Q*, *R*, Fallen conidia, each of which has put forth one two-celled adhesive outgrowth, *a*, from its proximal cell, and another, *b*, from its distal cell. *S*, Fallen triseptate conidium which has given rise directly to three sessile adhesive outgrowths, *a–c*, besides putting forth at its proximal end a stout hypha that bears on the side a unicellular adhesive process, *d*, and at the tip has a two-celled adhesive part, *e*. *T*, Fallen conidium which at one end has put forth directly a sessile adhesive outgrowth, *a*, shown as viewed endwise, and at the other end has produced a stout hypha bearing two bicellular adhesive outgrowths, *b* and *c*.

FIG. 7. *Dactylella cionopaga*, drawn to a uniform magnification with the aid of a camera lucida; × 500 throughout. *A*, Portion of mycelial hypha bearing a two-celled columnar adhesive outgrowth, *a*, and a three-celled columnar adhesive outgrowth, *b*. *B*, Portion of mycelial hypha whereon are borne a unicellular adhesive outgrowth, *a*, a two-celled columnar adhesive outgrowth, *b*, and a six-celled adhesive arch, *c*. *C*, Portion of prostrate hypha with a five-celled adhesive arch formed by distal union of two outgrowths. *D*, Portion of prostrate hypha bearing in addition to an inactive two-celled adhesive outgrowth, *a*, two other adhesive outgrowths, *b* and *c*, that have been operative in capturing a slender eelworm (possibly referable to *Plectus* sp.) and in intruding globose infective bodies from which assimilative hyphae have been extended through the animal. *E*, Portion of prostrate hypha with a two-celled adhesive outgrowth on whose distal cell a well developed eelworm is held affixed by means of a visible cushion of glutinous material; from the distal cell a globose infective body has been intruded that has given rise to growing assimilative hyphae. *F*, Portion of prostrate hypha with a two-celled adhesive outgrowth whose proximal segment is holding captive a slender eelworm; from a globose infective body intruded into the animal assimilative hyphae are being extended through its fleshy body. *G*, Portion of prostrate hypha with a two-celled adhesive outgrowth to whose basal segment a slender eelworm is held affixed; from the globose infective body intruded by the fungus a single assimilative hypha has been extended lengthwise through the animal's body. *H*, Portion of hypha bearing a three-celled adhesive outgrowth, whose proximal and distal segments have both been operative in capturing a slender eelworm; each of the two segments has

intruded a globose infective body from which one hypha is being extended forward and another backward through the animal's body. *I*, Prostrate hypha with a conidiophore from which a single conidium has been abjointed. *J*, Portion of prostrate hypha with an erect conidiophore, *a*, on which were produced the two conidia *b* and *c*, each of them shown with its basal end close to the position where it had been attached. *K*, Detached conidia showing usual variations in size and shape, together with variations in septation as follows: *a*, two septa present; *b–i*, three cross-walls present; *j–r*, four cross-walls present in symmetrical arrangement, two small cells being placed above and two small cells being placed below a large median cell; *s*, *t*, four cross-walls present, with the largest cell placed in penultimate position; *u–w*, five cross-walls present. *L*, Detached conidium that has put forth a unicellular adhesive outgrowth, *a*, from its basal cell. *M*, Detached conidium that has put forth one two-celled adhesive outgrowth, *a*, directly from its apical cell, and has extended from its tip a short stout hypha whereon is borne perpendicularly another two-celled adhesive outgrowth, *b*. *N*, Detached conidium that has put forth a two-celled adhesive outgrowth, *a*, from its tip, and from its basal cell has given off a unicellular adhesive outgrowth, *b*, as well as a four-celled adhesive outgrowth, *c*.

FIG. 8. *Dactylaria cudermata*, as found developing in nematode-infested agar plate cultures; drawn to a uniform magnification with the aid of a camera lucida; × 500 throughout. *A–D*, Portions of mycelial filaments, each bearing an adhesive hyphal network. *E*, Wide mycelial filament bearing a hyphal network, which, after being operative in capturing a sharp-tailed eelworm, *a*, intruded a globose infective body that has given rise to assimilative hyphae extending lengthwise through the fleshy interior; *b*, portion of the empty integument of a nematode that had been captured and depleted of its substance earlier; *c*, fusion of two assimilative hyphae. *F*, Portion of prostrate hypha with an erect conidiophore, *a*, from the tip of which has been abjointed a single conidium, *b*; the conidiophore being shown, from lack of space, in three parts whose proper connection is indicated by broken lines. *G*, Small portion of mycelial hypha with the proximal portion of a fallen conidiophore from which has been sent up a secondary conidiophore, *a*, that produced at its tip the conidium *b*; the conidiophore is shown, from lack of space, in three parts whose proper connection is indicated by broken lines. *H*, Detached mature conidia, *a–t*, showing usual variations in size and shape. *I*, Detached conidium that has sent up two slender aerial hyphae, *a* and *b*, from its parabasal cell, and one similarly slender aerial hypha, *c*, from its apical cell.

FIG. 9. *Dactylaria cudermata*, as found developing in nematode-infested agar plate cultures; drawn to a uniform magnification with the aid of a camera lucida; × 500 throughout. *A*, Portion of stout mycelial filament bearing a small adhesive network consisting of only two meshes. *B*, Portion of stout mycelial filament bearing a more extensive adhesive network. *C*, Successive stages in the closure of an adhesive hyphal bail: *a*, portion of mycelial filament bearing a completed hyphal bail to which a second hyphal bail is being added; as the elongating recurved uniseptate branch approaches distally the hyphal bail from which it originated the segment toward which its tip is directed extends a protuberance to meet it; *b*, same predacious ap-

paratus 40 minutes later than in *a*, the protuberance and the tip of the recurving branch now being in contact; *c*, same predacious apparatus 20 minutes later than in *b*, showing fusion of the protuberance and the recurved branch; *d*, same predacious apparatus 2½ hours later than in *c*, showing one septum formed near place of union and another septum formed about 20 μ backward, so that the completed hyphal bail is divided into three segments of about equal length. *D*, Portion of stout mycelial filament bearing an adhesive hyphal network in which a sharp-tailed eelworm (possibly referable to *Rhabditis* sp.) has been captured; the network has intruded three globose infective bodies from which assimilative hyphae, numbering six in all, are growing lengthwise through the animal. *E*, Portion of prostrate hypha bearing an erect conidiophore, *a*, that bore three conidia, one having been abjointed from the axial tip, *b*, another from the tip of a primary branch, *c*, and a third from the tip of a secondary branch, *d*; the conidiophore being shown, from lack of space, in three parts whose proper connection is indicated by broken lines. *F*, Portion of prostrate hypha with an erect conidiophore, *a*, that bore four conidia, one having been abjointed from the axial tip, *b*, two from tips of primary branches, *c* and *d*, and a fourth from the tip of a secondary branch, *e*; the conidiophore being shown, from lack of space, in three parts whose proper connection is indicated by broken lines. *G*, Mature detached conidia, *a–y*, showing usual variations in size and shape. *H*, Empty envelope of conidium that has yielded its contents to the mycelial hypha with which it is fused.

FIG. 10. Drawn to a uniform magnification with the aid of a camera lucida; × 500 throughout. *A–F*, *Dactylaria eudermata* as found in nematode-infested agar plate cultures: *A*, Portion of mycelium bearing an adhesive network in which a sharp-tailed eelworm (possibly referable to *Rhabditis* sp.) has been captured; a globose infective body is being intruded into the living captive. *B*, Portion of hypha bearing an adhesive network in which a sharp-tailed eelworm has been captured; from the single large bulbous infective body intruded by the fungus, assimilative hyphae have been extended lengthwise through the animal. *C*, Portion of prostrate hypha whereon is borne a conidiophore, *a*, from the tip of which a single conidium, *b*, has been abjointed; the conidiophore being shown, from lack of space, in three parts whose proper connection is indicated by broken lines. *D*, Detached conidia, most of them (*a–e*) divided by three cross-walls, but one (*f*) containing only two cross-walls. *E*, Conidium germinating by production of two germ tubes, one being extended from the basal end, the other arising laterally from the parabasal segment. *F*, Conidium that after fusing with a mycelial hypha has been emptied through transfer of its contents to the mycelium. *G–M*, *Dactylaria eudermata* as found in pure culture on maizemeal agar: *G*, Detached conidia, showing variations in size and shape, together with variations in number and position of septa as follows: *a–c*, three septa present, with the largest cell placed in penultimate position as in most conidia produced in nematode-infested cultures; *d–m*, two septa present, with the largest cell being terminal in position; *n–q*, one septum present, with the larger cell being in distal position. *H*, Conidium that has put forth an aerial hypha from its small parabasal cell. *I*, Conidium that has germinated by extending a germ tube from its minute basal segment. *J*, Conidium germinating by

putting forth one germ tube from its small basal cell, another from its slightly larger parabasal cell, and a third from its small apical cell. *K*, Conidium germinating by putting forth one germ tube from its rather small parabasal cell, and another from its small apical cell. *L*, Conidium that after becoming fused with a mycelial hypha, was emptied by transfer of contents to the mycelium. *M*, Conidium that after becoming fused with a mycelial hypha, has been nearly emptied through transfer of contents to the mycelium. *N–P*, Conidial apparatus, presumably belonging to *Dactylaria cudermata*, that was found in a nematode-infested culture: *N*, Fallen conidium from which have arisen two conidiophores, *a* and *b*, each bearing three unicellular secondary conidia. *O*, Tip of a conidiophore sent up from a fallen conidium resembling that shown in *N*; from this tip and its lateral prolongation were abjointed seven unicellular secondary conidia. *P*, Detached unicellular secondary conidia, *a–c*.

FIG. 11. *Dactylaria haptotyla*, as found developing in nematode-infected cultures; drawn to a uniform magnification with the aid of a camera lucida; × 500 throughout. *A*, Portions of mycelial hyphae, each bearing a predacious organ consisting of a globose adhesive cell and a unicellular (*a*, *b*) or a bicellular (*c–e*) stalk. *B*, Portion of hypha with two predacious organs, *a* and *b*, both having two-celled stalks. *C*, Portion of mycelium with four predacious organs, *a–d*, each having a two-celled stalk. *D*, Portion of hypha with a predacious organ whose stalk, on repeated elongation, bore successively three adhesive cells, *a–c*, whereof the two formed earliest, *a* and *b*, have degenerated. *E*, Portion of hypha bearing besides the inactive predacious organ *a*, two other predacious organs that have operated jointly in capturing a nematode (*Panagrolaimus* sp.) and have each intruded a globose infective body from which assimilative hyphae are being extended; one of these active predacious organs is attached to the mycelium not only by its original stalk, *b*, but also by a supplementary connection, *c*, while the other organ seems attached only by a supplementary connection, *d*. *F*, Portion of mycelium with a predacious organ, *a*, that has been operative in capturing a small nematode (*Panagrolaimus* sp.) and has intruded a globose infective body from which assimilative hyphae are being extended. *G*, Portion of mycelium with a predacious organ that has been operative in capturing a nematode (*Panagrolaimus* sp.) and has intruded into the captive a globose infective body from which assimilative hyphae are being extended; the distal segment of the bicellular stalk, *a*, originally supporting the adhesive cell, having been damaged, a supplementary connection, *b*, has been formed uniting the proximal segment of the stalk with the adhesive cell. *H*, Portion of mycelium with a predacious organ that has been operative in capturing a nematode (*Panagrolaimus* sp.) and has intruded a globose infective body from which assimilative hyphae are being extended; the adhesive cell here is found attached not only by its original stalk, *a*, but also by a supplementary stalk, *b*. *I*, Portion of prostrate hypha with an erect conidiophore, *a*, whereon is borne a conidium, *b*. *J*, Portion of prostrate hypha with an erect conidiophore, *a*, bearing one conidium, *b*, on its axial tip, and another conidium, *c*, on the tip of a distal spur. *K*, Portion of prostrate hypha with an erect conidiophore from which a single conidium has been abjointed. *L*, Detached conidia, *a–z*, showing usual variations in size and shape; all containing four cross-walls,

with the middle cell, except in *z*, exceeding the other cells in size. *M*, Detached triseptate conidium that has put forth an aerial hypha. *N*, *O*, Detached quadriseptate conidia which have each put forth an aerial hypha. *P*, Detached quinqueseptate conidium that has put forth an aerial hypha. *Q*, Detached quadriseptate conidium that has put forth two aerial hyphae. *R*, Two conidia, *a* and *b*, that have become fused by means of an anastomosing connection.

FIG. 12. *Dactylaria haptotyla*, as found developing in nematode-infested cultures; drawn to a uniform magnification with the aid of a camera lucida; ×500 throughout. *A*, Portions of mycelial hyphae, each bearing a predacious organ, the adhesive cell of which in some instances, *a–d*, is supported on a unicellular stalk, and in other instances, *e–k*, is supported on a bicellular stalk. *B*, Portion of mycelial hypha with two predacious organs, *a* and *b*. *C*, Portion of hypha bearing a young predacious organ, *a*, and two fully formed predacious organs, *b* and *c*; the stalk in *b* is commonplace in being composed of two segments, whereas that of *c* is exceptional in consisting of three segments. *D*, Portion of hypha bearing besides an inactive predacious organ, *a*, another predacious organ that has operated in capturing a nematode (*Panagrolaimus* sp.) and has begun intruding a globose infective body into the animal; the adhesive cell of the active organ is supported by the original stalk, *b*, and also by the supplementary hyphal connection, *c*. *E*, Portion of hypha with an adhesive organ, *a*, that has operated in capture of a nematode (*Panagrolaimus* sp.) and has intruded into the animal a globose infective body from which assimilative hyphae are being extended; the adhesive cell here is connected with the parent hypha only by its original stalk. *F*, Portion of hypha with a predacious organ that has operated in capturing a nematode (*Panagrolaimus* sp.) and has intruded a small infective body from which assimilative hyphae are being extended; the adhesive cell here is attached to the parent hypha by its original two-celled stalk, *a*, and also by a supplementary connection, *b*. *G*, Portion of hypha with a predacious organ that has operated in capture of a nematode (*Panagrolaimus* sp.) and has intruded a globose infective body from which assimilative hyphae have been extended lengthwise through the animal; the adhesive cell having degenerated, its interior is in part occupied by a prolongation from the supplementary connection, *a*, which provides now the only communication between the assimilative hyphae and the external mycelium; at *b* is shown an anastomosis of an assimilative hypha with the tip of one of its branches. *H*, Portion of hypha with two predacious organs that have operated jointly in capturing a nematode (*Panagrolaimus* sp.), and have each intruded a globose infective body from which assimilative hyphae have been extended; the stalk *a* originally suporting one of the adhesive cells having degenerated distally, a supplementary connection, *b*, has been formed between the adhesive cell and the uninjured proximal segment of the stalk; *c*, supplementary hyphal connection that replaces the stalk originally supporting the other adhesive cell; *d*, *e*, anastomoses of assimilative hyphae. *I*, Portion of prostrate hypha with two conidiophores, *a* and *b*, which support aloft three (*c–e*) and four (*f–i*) conidia, respectively. *J*, Portion of mycelium with an erect conidiophore from which three conidia have been abjointed. *K*, Detached conidia, showing usual variations in size and shape, together with variation in septa-

tion as follows: *a*, *b*, three septa present; *c–x*, four septa present; *y*, *z*, five septa present. *L–N*, Quadriseptate conidia that have each put forth an aerial hypha, as usually, from the parabasal cell.

FIG. 13. *Dactylaria sclerohypha*, as found developing in nematode-infested agar plate cultures; drawn to a uniform magnification with the aid of a camera lucida; × 500 throughout. *A*, Portions of hyphae, *a–q*, each bearing a predacious organ with a two-celled stalk. *B*, Portion of hypha with two predacious organs, *a* and *b*, each having a two-celled stalk, though in *b* the proximal cell extends into the parent hypha. *C*, Portion of hypha with three predacious organs, two of them (*a*, *b*) having bicellular stalks, the other (*c*) having a unicellular stalk. *D*, Portion of mycelium bearing one predacious organ, *a*, with a two-celled stalk, and four other predacious organs, *b–e*, with unicellular stalks; *d* has been operative in capturing a nematode and has intruded a globose infective cell from which assimilative hyphae have been extended lengthwise through the animal; the predacious organ *e*, together with the two infective cells and the assimilative hyphae it extended into a captured nematode, is shown empty of protoplasmic contents. *E*, Portion of mycelium bearing two inactive predacious organs (*a* and *b*) with bicellular stalks, in addition to two predacious organs (*c* and *d*) that have each operated in capturing a separate eelworm and have each intruded into their captive a globose infective body from which assimilative hyphae have been extended; both *c* and *d* are attached mainly by supplementary connections; *e*, *f*, anastomoses of assimilative hyphae. *F*, Portion of hypha bearing a predacious organ with a unicellular stalk. *G*, Portion of hypha bearing one inactive predacious organ (*a*) with a unicellular stalk, and two predacious organs (*b* and *c*) that have each operated in capturing a separate nematode and have each intruded a globose infective body from which assimilative hyphae have been extended lengthwise within the animal; both active organs (*b* and *c*) show development of a supplementary hyphal connection; *d*, anastomosis of assimilative hyphae. *H*, Portion of hypha bearing an inactive predacious organ (*a*) with a unicellular stalk and also a predacious organ (*b*) that has operated in capturing a nematode and has intruded a globose infective body from which assimilative hyphae have invaded the animal lengthwise. *I*, *J*, Portions of mycelium each bearing a predacious organ with a two-celled stalk; the proximal cell in both instances extends into the parent hypha. *K*, Portion of mycelium bearing one predacious organ (*a*) with a unicellular stalk and another (*b*) with a long four-celled stalk. *M*, Portion of hypha bearing a predacious organ that has operated in capturing a nematode and has intruded a globose infective body from which assimilative hyphae have invaded the animal lengthwise; *a–c*, anastomoses of assimilative hyphae. *N*, Portion of mycelium bearing a predacious organ that has operated in capturing a nematode and has intruded a globose infective body from which assimilative hyphae have grown lengthwise in the animal; the empty distal portion of the two-celled stalk originally supporting the adhesive cell has been supplanted by a short supplementary hyphal connection. *O*, Portion of hypha with a predacious organ that after operating in the capture of a nematode has intruded a globose infective body from which assimilative hyphae have grown lengthwise through the animal; the

adhesive cell here is attached by its original two-celled stalk, *a*, and also by a supplementary connection, *b*.

FIG. 14. *Dactylaria sclerohypha*, as found developing in nematode-infested agar plate cultures; drawn to a uniform magnification with the aid of a camera lucida; × 500 throughout. *A*, Portion of prostrate mycelium from which has been sent up an erect conidiophore, *a*, bearing three conidia, *b–d*. *B*, Portion of mycelium with a prostrate hypha representing apparently a fallen conidiophore, *a*, which has sent up from a proximal segment an erect secondary conidiophore, *b*, bearing eight conidia, *c–j*, in loose capitate arrangement. *C*, Erect conidiophore bearing five sterigmata in rather close arrangement, from each of which a conidium has been abjointed; the branching hyphae at the base consist largely of proximal portions of older conidiophores that have fallen over on to the substratum. *D*, Prostrate mycelial hypha with proximal portions of both the fallen primary conidiophore *a*, and the fallen secondary conidiophore *b*; from the latter is shown arising a tertiary conidiophore, *c*, with five denuded sterigmata. *E*, *F*, Distal portions of denuded conidiophores provided with sterigmata in numbers of three and five, respectively. *G*, *H*, Random assortment of detached conidia, *a–z*, showing usual variations in size, shape, and septation. *I*, Detached conidium that has put forth a predacious organ from its distal end, and an erect aerial hypha from its parabasal cell. *J*, Detached conidium that has put forth a predacious organ from its median cell and an erect aerial hypha from its parabasal cell. *K*, Detached conidium that has put forth an erect aerial hypha from its parabasal cell. *L*, Two detached conidia, *a* and *b*, united through vegetative fusion, that have sent up the conidiophores *c* and *d*, respectively, which bear the conidia *e* and *f*, respectively. *M*, Cuticle of a captured nematode within which the assimilative hyphae and the globose infective body have become conspicuously indurated. *N*, Portion of an extensive branching hyphal system consisting mostly of somewhat thick-walled, enlarged, globose, indurated cells (or chlamydospores) with globuliferous contents.

FIG. 15. *Gonatobotrys simplex*, drawn to a uniform magnification with the aid of a camera lucida; × 500 throughout. *A*, Portion of prostrate hypha with an erect conidiophore bearing five conidial clusters *a–e*; drawn from material that developed on a tomato leaf in a moist chamber at 18° C. *B*, Denuded terminal portion of a conidiophore formed on a tomato leaf in a moist chamber at 18° C.; the conidiophore shows four noticeably swollen nodes, *a–d*, beset with truncate protuberances from which conidia have been abjointed. *C*, Denuded conidiophore with a swollen tip, *a*, bearing truncate protuberances from which conidia were abjointed; clusters of conidia likewise were abjointed from the swollen tips, *b* and *c*, of the two lateral branches; to the prostrate hypha from which the conidiophore arises is attached a globose unicellular branch, *d*, and a bicellular branch, *e*; drawn from a pure culture of the muskmelon strain on maizemeal agar; from lack of space the conidiophore is shown in three parts whose proper connection is indicated by broken lines. *D*, Portion of primary conidiophore, *a*, from which a secondary conidiophore, *b*, has been sent up; one denuded sporiferous node, *c*, is shown in the primary conidiophore, and another, *d*, in the secondary conidiophore; the swollen tip *e* of the secondary conidiophore is beset with conidia in a very early stage of development; drawn from a pure culture of the muskmelon

strain on maizemeal agar. *E*, Portion of primary conidiophore, *a*, from which a secondary conidiophore, *b*, has been sent up; a denuded sporiferous node, *c*, is shown in the primary conidiophore, and another, *d*, in the secondary conidiophore; the swollen tip, *e*, of the secondary conidiophore is beset with young growing conidia; produced on a tomato leaf in a moist chamber at 18° C. *F* (*a–z*), *G* (*a–o*), Detached conidia showing usual variations in size and shape; taken from a tomato leaf that had been kept in a moist chamber at 18° C. *H* (*a–z*), *I* (*a–h*), Detached conidia taken from a muskmelon leaf that had been kept in a moist chamber at 18° C. *J*, Detached conidia, *a–o*, as found in a pure maizemeal-agar plate culture of the muskmelon strain seven days after inoculation. *K*, Two uniseptate conidia, *a* and *b*, found among a multitude of non-septate conidia produced by the fungus on a tomato leaf in a moist chamber at 18° C. *L*, Portion of mycelium from a maizemeal-agar culture, showing three unicellular globose branches, *a–c*, and a bicellular branch, *d*. *M*, Portion of mycelium from a maizemeal-agar culture, showing ten unicellular globose or ellipsoidal branches, *a–j*, and two bicellular branches, *k* and *l*. *N*, Portion of mycelium from a maizemeal-agar culture, showing five unicellular globose or ellipsoidal branches, *a–e*, and a bicellular branch, *f*. *O*, Portion of mycelium from a maizemeal-agar culture, showing four unicellular globose branches, *a–d*, borne laterally on a longish hypha, and two others that are borne separately on a unicellular branch, *e*, and a bicellular branch, *f*. *P*, Portion of mycelium from a maizemeal-agar culture, showing eight short unicellular branches, *a–h*, and a somewhat longer unicellular branch, *i*.

NEW OR NOTEWORTHY FUNGI FROM MT. RAINIER NATIONAL PARK*

ALEXANDER H. SMITH AND DANIEL E. STUNTZ

(WITH 21 FIGURES)

As part of a general program involving a manual on the gill-fungi of our western United States, which has occupied the senior author for a number of years, the collecting season of 1948 was spent at Mt. Rainier National Park. The personnel of the expedition included Mr. Henry Imshaug, the senior author's assistant, and Mr. Emory Simmons, who was collecting Pyrenomycetes for Prof. L. E. Wehmeyer. The junior author joined the party early in July. Laboratory space and living quarters at Longmire were provided for us by the Park authorities. The entire personnel of the expedition is greatly indebted to Mr. John C. Preston, Superintendent, to Mr. Russell Grater, Naturalist, to Mr. Merlin K. Potts, Assistant Naturalist, as well as to Dr. E. T. Bodenberg, Ranger Naturalist, for the facilities received and also for many helpful suggestions and courtesies which added greatly to the efficiency with which we were able to carry out our studies.

We began work during the last week in June and continued until the onset of cold weather and snow at the end of September. The season was an unusual one in that, because of frequent rains, the woods remained moist all summer. Fleshy fungi were fairly abundant until near the middle of September, at which time, because of a short period without rain, the production of fruiting bodies slackened appreciably. Although ample rain came after that date, relatively cool temperatures, apparently, retarded any further development of fleshy fungi. Although the fall mushroom

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flora, characteristically the most prolific one of any season in this area, was exceptionally poor, the summer flora was exceptionally good. Since the summer agaric flora of the Cascades as a whole is the most difficult to obtain, we felt very fortunate indeed. Without a doubt these circumstances are largely responsible for the considerable number of new and interesting fungi found in the area. Both of us have collected extensively in the Pacific Northwest, but our activities have been limited mostly to spring and fall collecting.

The area which we covered within the Park was relatively limited since the richness of many spots required that they be visited repeatedly. The lower Nisqually River, below the junction of Kautz Creek, was one such area, but the lower Tahoma Creek Valley, from where the creek crosses the road to Longmire on up to the Tahoma Creek Forest Camp, was the most outstanding. A majority of our collections came from these two areas. A great deal of time was spent working the higher country, but the fungous flora which came out as the snow melted was limited in extent and persisted until snow came again in the fall. We believe that this represented an unusual fruiting pattern which was dictated by unusual weather conditions. Only one trip was made up the Carbon River Valley, and no collecting was done between the Carbon River and Round Pass. Mr. Imshaug spent the early part of the season collecting at and above Ohanapecosh Hot Springs. From this it can be readily seen that in spite of making over 3500 collections of fleshy fungi, an overall picture of the Park flora in this group was not obtained.

Because we were interested particularly in *Psathyrella* and *Inocybe* respectively a special study was made of each genus. Fortunately both genera were well represented in the flora. The following species represent a few of the more outstanding discoveries. Additional data on fungi in other genera will be published by us at a later date, or by those to whom material has been sent for critical study.

Color names cited from Ridgway (**1912**) are enclosed within quotation marks. Those cited from Maerz and Paul (**1930**) are indicated as follows: raw umber 15A12, etc.

Clitocybe subvelosa sp. nov. *Fig. 21c*

Pileus 3–9 (10) cm. latus, subplanus, margine involutus, demum subinfundibuliformis, viscidus, aurantio-cinnamomeus vel incarnato-cinnamomeus, odor farinaceus, sapor praerancido-farinaceus; lamellae avellaneae vel incarnato-avellaneae demum subfuligineae, confertae, late decurrentes; stipes 6–9 cm. longus, 8–15 mm. crassus, aequalis vel deorsum clavatus, solidus, intus avellaneus, externe "pale vinaceous fawn" vel "vinaceous buff" et fibrillosus, sursum fibrilloso-annulatus; sporae 8–10.5 × 5–5.5 μ, ovatae vel subellipticae.

Pileus 3–9 (10) cm. broad, disc flat and margin inrolled at first, expanding to shallowly depressed on disc and with spreading or arched margin, surface viscid, toward the margin with streaks of agglutinated fibrils, edge at first fringed with veil remnants, disc glabrous and in some watery-spotted, color "orange-cinnamon" or darker at first, margin "light pinkish cinnamon" colors duller in age; flesh "pale pinkish buff" except near gills where it is watery avellaneous, odor farinaceous, taste intensely bitter-farinaceous (very rancid); lamellae "avellaneous" to "vinaceous buff" young, becoming "wood brown" to subfuligineous in age, close to crowded, broad, decurrent, not separable, often forked, edges even; stipe 6–9 cm. long, 8–15 mm. in diam., equal to clavate, solid, more or less avellaneous in cortex, surface "pale vinaceous fawn" to "vinaceous buff" and appressed fibrillose, cottony at base, with a thin fibrillose annular zone of partial veil fibrils near apex.

Spores 8–10 × 5–5.5 μ, white in deposit, ovate pointed at maturity, nonamyloid, smooth; basidia 36–40 × 5–7 μ, four-spored, hyaline in KOH, yellow in iodine; pleurocystidia and cheilocystidia none differentiated; gill trama (as seen in sections of fresh material) distinctly of interwoven hyaline filaments; pileus trama homogeneous beneath a distinct gelatinous pellicle composed of narrow (2.5–4 μ) orange-brown hyphae as seen in water mounts of fresh material, hyaline revived in KOH; clamp connections present.

Habit, habitat and distribution: gregarious under alder and conifers, lower Tahoma Creek, September 12. Coll. by D. E. Stuntz (Sm. *31182*—**type**).

Discussion: The intense taste, incarnate-orange color, viscid cap, fibrillose veil, and the change in the color of the gills from youth to age amply distinguish this fungus from other species in the genus. It appears to be most closely related to *C. gomphidioides* in all characters except the presence of a distinct fibrillose veil and in the shape and size of the spores. The differences in the spores are shown (FIG. 21*c*, *d*). Drawings for both species

were made from the types. My first impression of this fungus was that it belonged in *C. gomphidioides* and that I had failed to note the veil in the Olympic specimens. However, a critical comparison of the dried specimens and the photographs of each made at the time of collection shows my first impression to have been false. When one considers other instances of parallel species, largely distinguished by the presence of a veil in one and its absence in the other, it is evident that the logical procedure is to regard *C. subvelosa* as distinct from *C. gomphidioides*. It should be kept in mind that for years it was questioned whether or not *Armillaria mellea* and *Clitocybe tabescens* were distinct. Rhoades (**1945**) has clearly established that they are. Another parallel situation is that of *Hygrophorus erubescens* (veil-less) and *Hygrophorus purpurascens* (with a fibrillose veil). Since both *C. subvelosa* and *C. gomphidioides* bear a striking resemblance to species of *Hygrophorus* possessing divergent gill trama, a careful study of the arrangement of the hyphae of the gill trama in *C. subvelosa* was made from both young and old caps in the fresh condition. In both stages it was found to be interwoven.

Cortinarius rainierensis sp. nov. *Fig.* 1

Pileus 3–8 cm. latus, obtusus vel conicus demum campanulatus vel subplanus, siccus, squamulosus, "russet" vel "tawny," saepe "Sanford's brown"; odor raphanaceus; lamellae fulvae demum subferrugineae, latae, subdistantes, venosae; stipes 5–8 (10) cm. longus, 10–12 (15) mm. crassus, farctus, deorsum demum obscure ferrugineus, fibrilloso-annulatus; sporae 9–11 × 6.5–8 μ, late ovatae.

Pileus 3–8 cm. broad, obtuse to convex, expanding to conic campanulate, conic umbonate, obtusely campanulate or broadly convex, sometimes nearly plane in age, surface dry, innately fibrillose-squamulose over disc or nearly to margin, "russet" on disc and "tawny" over marginal area, when fruiting luxuriantly evenly rich tawny to "Sanford's brown," unchanging or merely slightly darker in extreme age; flesh rather thick in disc, thin away from the stipe, pale buff, odor of radish, taste similar but very slight; lamellae "ochraceous tawny" young, soon "tawny" and remaining so, broad, subdistant, adnexed, thick and intervenose, edges even; stipe 5–8 (10) cm. long, 10–12 (15) mm. thick, stuffed with a pale buff pith, cortex pale to dark tawny, darkening in base to Sanford's brown or darker where handled, mycelium white.

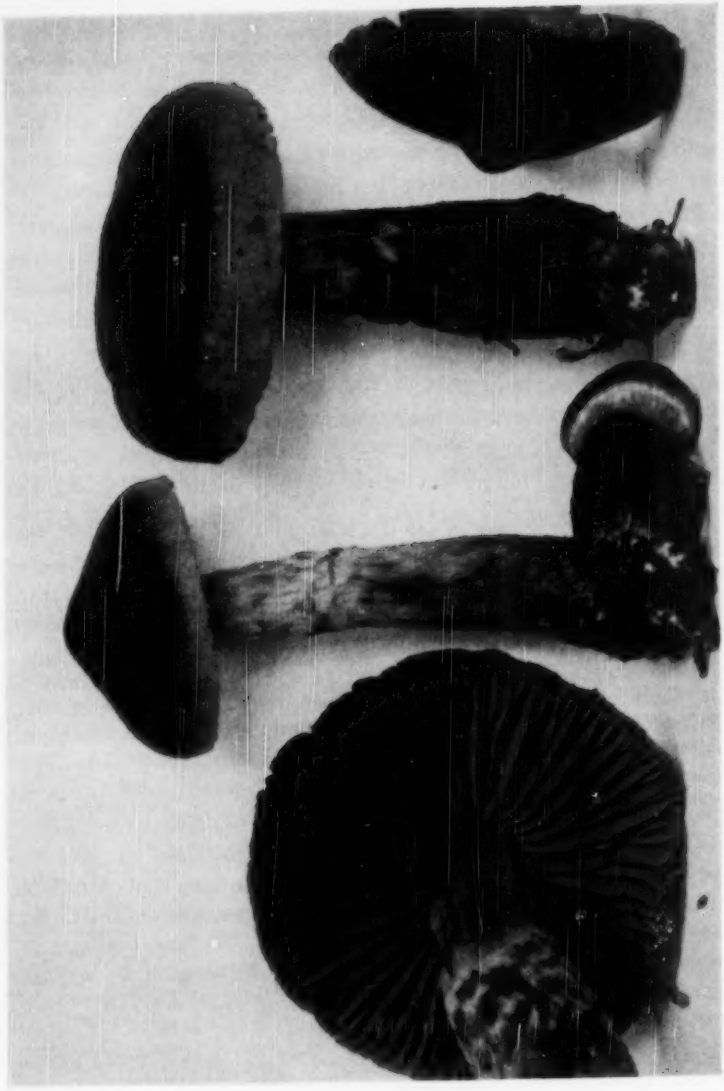

FIG. 1. *Cortinarius rainierensis*, ×1. Photo A. H. Smith

Spores dark rusty brown in KOH, 9–11 × 6.5–8 μ, broadly ovate, surface roughened; basidia four-spored, 38–52 × 8–10 μ, hyaline in KOH (many brownish collapsed basidia also present); pleurocystidia none; cheilocystidia basidia-like but smaller, 30–40 × 5–8 μ, narrowly clavate to subcylindric and hyaline or nearly so; gill trama parallel or nearly so, yellowish (in thin sections) to rusty reddish (in thick sections) in KOH, pileus characterized by fascicles of rusty brown to yellow-brown (in KOH) hyphae projecting to form squamules, pigment incrusted on the wall but deposits not heavy, clamp connections present.

Habit, habitat and distribution: Scattered to gregarious on humus in virgin mossy Douglas fir-hemlock forests. August 18, lower Tahoma Creek (Sm. *30281*—**type**). The first collection to come to our attention was one by Stuntz (Sm. *29504*) at Barlow Pass in the Washington Cascades July 26, 1948. Numerous collections, however, were made in the Park as follows: Imshaug *2126*, Nisqually River below Longmire, August 30; Imshaug & Smith (Sm. *29806*), lower Kautz Creek, August 4; Sm. *30535*, lower Tahoma Creek, August 23; Sm. *31083*, September 9; Sm. *31632*, Longmire, September 22.

Discussion: This is a striking species and one which can be recognized at a glance. At maturity it is practically unicolorous. The color and dry scaly cap are sufficient for field identification. In the herbarium it is macroscopically indistinguishable from *C. distans* var. *olympianus* Smith. However, the latter has distinctly narrower spores, 5–6 μ as contrasted to 6.5–8 μ, which makes collections easily recognizable when dried. In the fresh condition *C. distans* var. *olympianus* is hygrophanous and when moist has a faintly striatulate margin. *C. rainierensis* appears to be related to *C. annulatus* by the odor of radish, scaly cap and fibrillose ring which is usually present on the stipe, but differs in the much larger spores. Among the European species it appears close to *C. tophaceus* but differs in the rusty brown rather than ocher-yellow color. *C. fulvaureus* Henry and *C. speciosus* Favre, two recently described species, appear to be somewhat similar in stature. However, the former differs in its spore characters and the latter in its much more brilliant colors as clearly indicated by Favre (**1948**) both in his description and illustration.

CORTINARIUS SUBTORTUS Fries *Figs. 2; 3a*

Pileus 3.5–5 cm. broad, buttons broadly convex to flattened and the margin inrolled, expanding to broadly convex or nearly plane, glabrous, glutinous to viscid but soon dry and shining, some with agglutinated fibrils along margin, variously colored "Isabella color" to "honey yellow" and finally near "Sayal brown" or mottled "dark olive-buff" on an "Isabella color" background, some "light brownish olive" over all, occasionally splashed with fulvous in age; flesh mottled "snuff brown" and "buffy brown," becoming honey-yellow or more fulvous in age, taste mild to slightly bitterish or with bitterish aftertaste, odor faintly pungent but hardly distinctive; lamellae "citrine drab" to "buffy brown," in some "brownish olive," "light brownish olive" or "Isabella color," slowly changing to cinnamon brown or more fulvous (near "raw sienna"), depressed-adnate to broadly adnate or subdecurrent, narrow and intervenose but finally moderately broad, edges even; stipe short, 3–5 cm. long, (4) 6–10 (15) mm. thick, clavate, becoming subequal, stuffed with a white pith, cortex avellaneous, surface olive-buff from a dense coating of fibrils, with a median to superior fibrillose zone and at times the sheath breaking up into obscure zones, silky and sordid olivaceous above at first, in age paler and more yellowish (near "honey color"), a few (in Sm. *27980*) with an obscure violaceous tinge in apex.

Spores 7–9 × 6–7 μ, broadly ovate-pointed to subglobose, rusty brown in KOH, slightly roughened; basidia 27–33 × 7–8 μ, four-spored, yellowish in KOH; pleurocystidia abundant, 50–80 (100) × 8–10 μ, in water mounts of fresh material often with an incrusting sheath, smooth as revived in KOH, hyaline and thin-walled, narrowly ventricose to subcylindric or neck tapered, wavy, and with acute apices; cheilocystidia similar to pleurocystidia but shorter; gill trama parallel, pale yellow in KOH; cuticle of cap a gelatinous pellicle, the hyphae yellowish.

Habit, habitat and distribution: Gregarious to subcespitose under conifers, especially around very decayed stumps and logs, lower Nisqually above junction with Kautz Creek, and along highway near Tumtum Mt., Sept. 22 (Sm. *31608*), and Sept. 23 (Sm. *31634*). The senior author first collected this species on the east fork of the Salmon River, Mt. Hood, Oregon, at an elevation of about 4500 ft., Oct. 20, 1947 (Sm. *27980*).

Discussion: This species is very distinctive in the genus *Cortinarius* because of the presence of the greatly elongated pleuro-

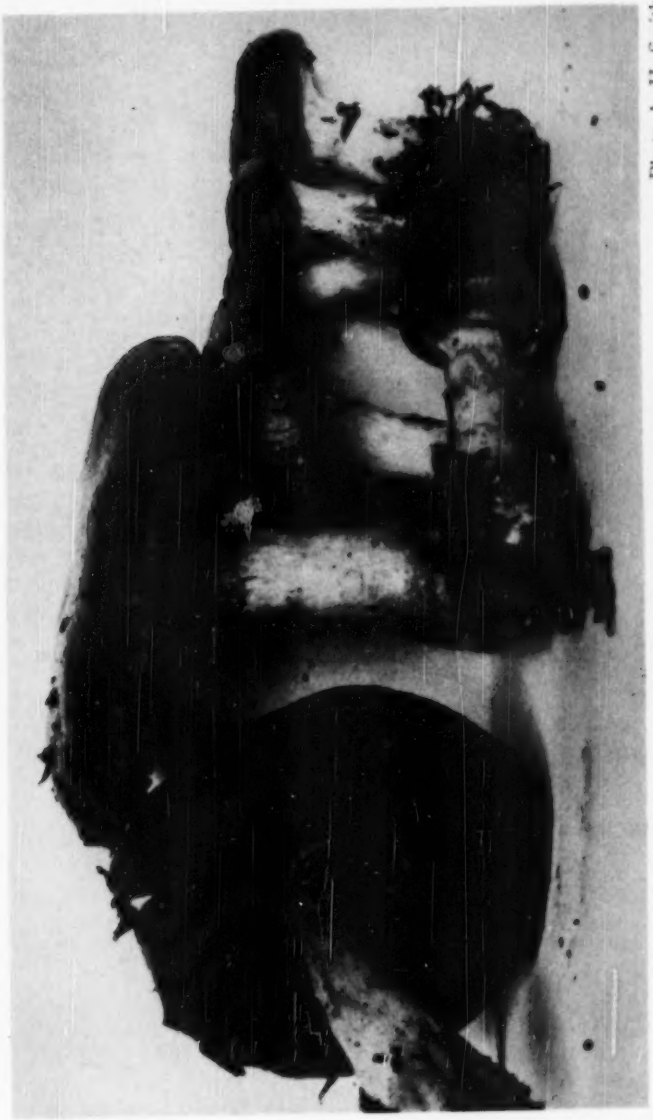

Photo A. H. Smith

FIG. 2. *Cortinarius subtortus.*

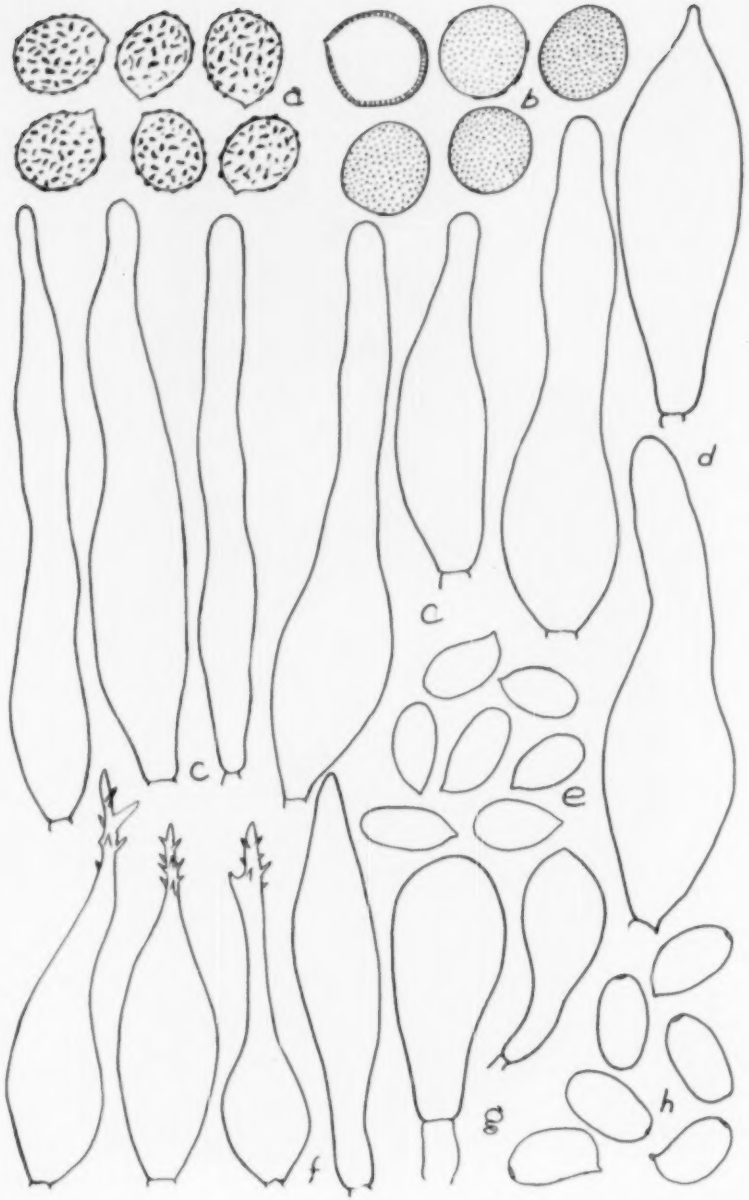

FIG. 3. Spores and cystidia of *Cortinarius*, *Mycena* and *Psathyrella*.

cystidia and a general resemblance to *C. infractus* in color. Maire et Kühner (**1935**) reported on this species as they found it in two different localities. Maire's collection was characterized by violaceous tints in stipe and gills. Coll. Sm. *27980* from Mt. Hood showed a slight tendency toward violaceous in the apex of the stipe. However, in the only extensive fruiting of this species we have observed here in North America, namely that made near Tumtum Mt. in the Park, the colors were more like those of Kühner's collection.

INOCYBE ARMORICANA Heim *Figs.* 4; 5*a*, *b*

Pileus 2.5–9 cm. broad, at first conic-campanulate, becoming expanded with a large prominent mammiform umbo, the margin soon plane and then finally a little uplifted; surface silky-smooth, lubricous when moist, drying silky-shining, often with a thin webby pallid coating of surface fibrils persisting as pallid patches at the center, toward the margin becoming more or less long-rimose, and at times regularly and conspicuously so, color at first "cadmium yellow" to "raw sienna," becoming "tawny" to chamois 11I5, 12E8, toast 13F8, or 13G9 at the margin; context 2–4 mm. thick off the disc, firm, white, unchanging on exposure, odor at first of green corn, then changing and becoming rather sweet and aromatic but still with an admixture of green corn; lamellae adnexed to almost free, broadly and shallowly rounded at the stipe, pointed at the margin, ventricose, widest toward the margin, 5–7 mm. broad, rather close, color white at first, becoming grayish olive (Arizona 13E6 to 14E6); stipe 4–12 cm. long, 5–13 mm. thick, terete or flattened, equal or in some a little incrassated downward, solid, the context firm and white to pallid yellowish, unchanging on exposure, usually becoming yellowish at the surface in age, base abruptly bulbous (submarginate) to clavate-bulbous and 13–20 mm. thick, surface satiny, glabrous to very slightly fibrillose-streaked, longitudinally striate, sometimes pruinate at the apex, color at first white, becoming pallid yellow all over, in age more or less flushed with darker shades of yellowish brown such as cinnamon 12E7 or 12F7, sometimes also with a subtle vinaceous cast.

Spores 7–8.5 (10) × 5–6 μ, subreniform, smooth; pleurocystidia lacking; cheilocystidia 45–67 × 20–25 μ, broadly clavate.

Under conifers, Nisqually River at Southwest entrance to Park, A. H. Smith, July 28; lower Tahoma Creek, Smith and H. Imshaug, August 2 (St. *3813*); same locality, Smith, Imshaug

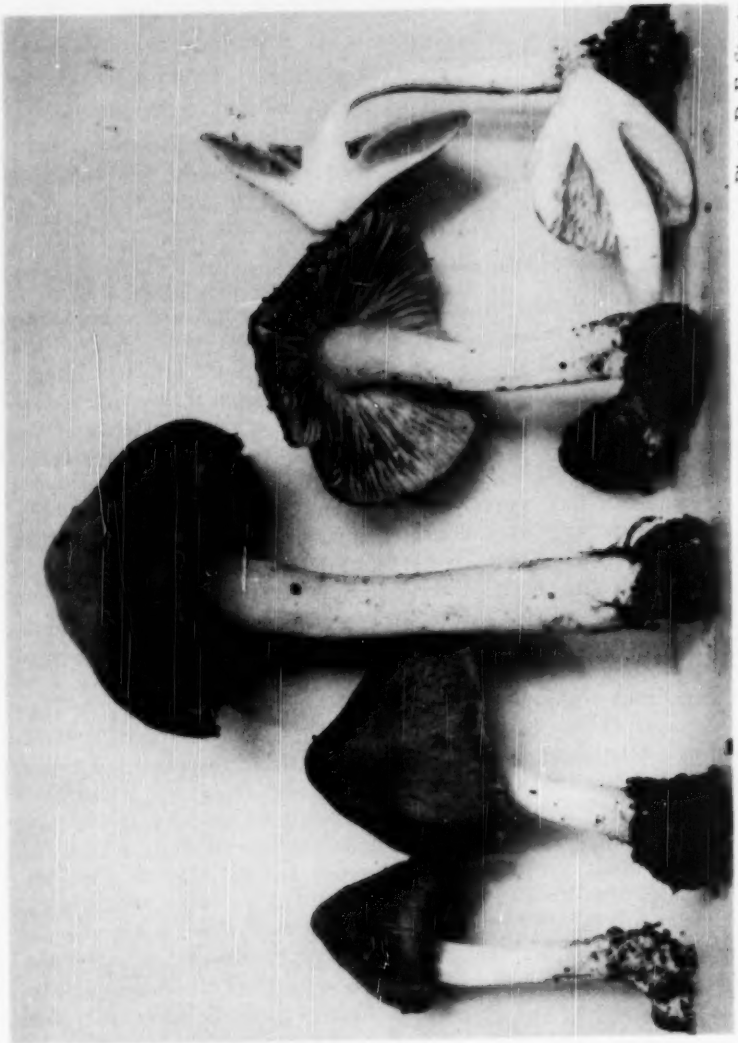

FIG. 4. *Inocybe armoricana*, ×1. Photo D. E. Stuntz

and Grace Howard, August 3 (St. *3880*); same locality, August 9 (St. *3910*); lower Tahoma Creek, at the beaver dam, Elizabeth Knowles and D. E. Stuntz, August 15 (St. *3975, 3997*); same locality, August 21 (St. *4098*), September 12 (St. *4427*), Smith and Stuntz, September 14 (St. *4498*), Smith, Knowles, Margaret McKenny and Stuntz, September 19 (St. *4620*), September 21 (St. *4682*), September 22 (St. *4758*).

This is the first report of *Inocybe armoricana* Heim from North America. It was the dominant species of *Inocybe* in the lower Tahoma Creek area in late July and early August, and continued to appear sporadically in that area throughout the remainder of the collecting season. Its smooth yellow-orange to tawny pileus, rather massive white to pallid yellow stipe, and peculiar complex odor are quite constant characteristics by which it can be easily recognized. Some of the specimens are more than twice as large as the maximum dimension given by Heim (**1931**, p. 295), and in all the collections the strong green corn odor was much less modified by the subsequent development of a sweet or aromatic component than is indicated by Heim. Both these discrepancies seem to fall within the range of variability to be expected in the species, however, and scarcely could be considered sufficient grounds for excluding the collections from *I. armoricana*. Heim indicated (**1931**, p. 297) a close relationship between this species and those related to *Inocybe jurana* Patouillard, especially *I. subrubescens* Atkinson. It might appear that an equally close or even closer relationship exists between *I. armoricana* and *I. sororia* Kauffman, since the principal differences between the two are those of pileus color and spore size, and the modification which the green corn odor of *I. armoricana* undergoes with the passage of time. Thus the *jurana* and the *fastigiata* species complexes may not be as unrelated as is commonly supposed.

INOCYBE FASTIGIATA (Fr.) Quél. f. ALPESTRIS Heim *Fig. 5c, d*

Pileus 4 cm. broad, 3.5 cm. high, conic-campanulate, sharply umbonate, with slightly incurved margin soon becoming long-rimose, color pale buff with an obscure tinge of tawny, near "cinnamon buff"; context about 2 mm. thick just off the disc, firm, white, unchanging on exposure, absolutely without odor; lamellae

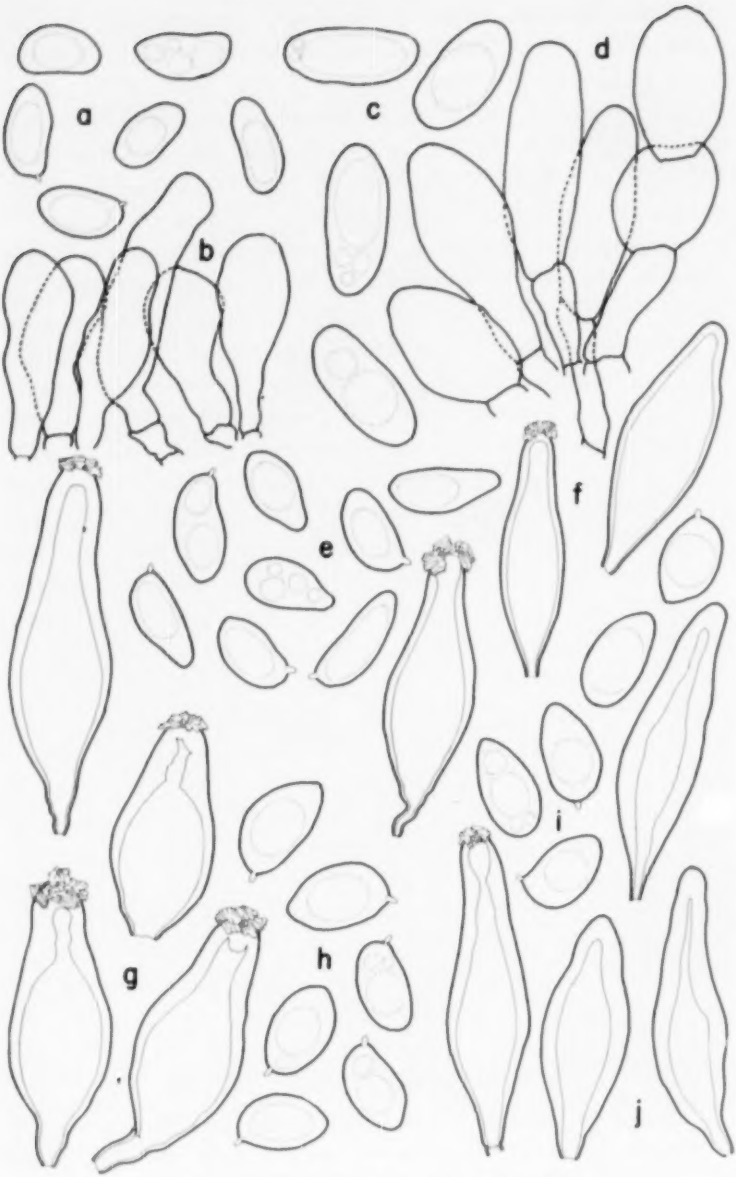

FIG. 5. Spores and cystidia of *Inocybe*.

adnate, plane both at stipe and margin, linear-oblong, narrow, 4 mm. broad, crowded, color pale grayish olive, with rather abundant brownish stains, the edges conspicuously white-fimbriate; stipe 9 cm. long, 6.5 mm. thick, terete, equal, flexuous, the base abruptly bulbous, 11 mm. in diameter, solid, context hard, white, unchanging on exposure, surface satiny, shining, sparsely white-fibrillose-streaked, color white, with a very faint tinge of yellow in some.

Spores 11.5–15 (16.5) × 6.5–7.5 μ, subcylindrical, occasionally subreniform; pleurocystidia lacking; cheilocystidia 36–37.5 × 16–26 μ, clavate, simple or often in short chains of two to three cells.

Under conifers, lower Tahoma Creek near the highway, A. H. Smith, July 29 (St. *3759*), and again in the same area, Smith and H. Imshaug, July 30 (St. *3796*).

Among the numerous varieties and forms of *Inocybe fastigiata* (Fr.) Quél. that occur in Washington, this one is easily recognized by its pallid yellowish buff pileus, long slender white stipe, and complete lack of any kind of an odor. It might be taken for a slender specimen of *I. sororia* Kauffm., but the lack of that species' characteristic strong green corn odor soon corrects that impression.

The use of the form name *alpestris* may require some explanation. It is fairly evident (see **1931**, p. 176, and the footnote on p. 392) that Heim did not really intend what he called "forme alpestris" to be given actual taxonomic status; yet he assigns varietal names to other collections (var. *arenicola*) some of which so closely resemble "forme alpestris" that one wonders why they were not included in it. The more luxuriant specimens, at least, of var. *arenicola*, are evidently more robust plants than "forme alpestris," are white rather than yellowish, and are said to have a detectable spermatic odor. Our collections are closer in these particulars to "forme alpestris" than to var. *arenicola*, and since they are readily distinguishable from typical *I. fastigiata*, the designation we have used for them seems justifiable.

INOCYBE GRISEO-LILACINA Lange *Figs. 5e, f; 6*

Pileus 1.5–3 cm. broad, obtusely campanulate, becoming expanded and plane or nearly so, with a rounded umbo, the margin persistently rounded, surface densely appressed-fibrillose, the cuticle becoming diffracted-scaly all over with furfuraceous squamules which are appressed to slightly recurved at the tip, color yellowish

buff on the umbo (walnut 12E6 to bamboo 13I7), elsewhere dingy yellowish gray (Long Beach 12B4 or polo tan 13C6), context about 2 mm. thick off the disc, pallid, unchanging on exposure, odor spermatic but not strong; lamellae adnexed, somewhat angularly sinuate, bluntly pointed at the margin, subventricose, 2–3 mm. broad, moderately close, color at first pallid with a flush of violaceous, becoming dingy grayish (Aloma 13C7), finally quite dark olivaceous umber (15E12); stipe 2.5–4 cm. long, 3–5.5 mm. thick, terete, equal, usually flexuous, the base rounded-truncate, not at all bulbous; solid, the context pallid within, dull lilac at the surface, unchanging on exposure, surface densely appressed-fibrillose, the

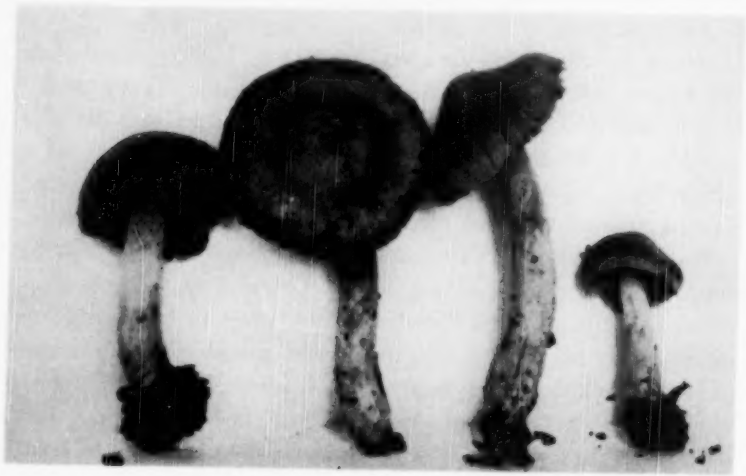

Photo D. E. Stuntz

FIG. 6. *Inocybe griseolilacina*, × 1.

fibrils in tiny pointed longitudinally oriented streaks especially below, the apex pruinate and almost glabrous, color pallid grayish buff (near beige soiree 11B4) at the base, lilac above (bisque 11A3 to tawny birch 13A6), but made to appear considerably duller because of the dense coating of surface fibrils.

Spores 8–9 (11) × 4.5–5.5 μ, inequilaterally amygdaliform; pleurocystidia 45–67 (80) × 14.5–18 μ, ventricose, with pedicel and slender neck, usually thick-walled; cheilocystidia of two kinds, some like the pleurocystidia in size and shape, the majority clavate to ovoid, thin-walled, 20–30 × 11–13.5 μ, densely clustered along the gill edge.

Under conifers, lower Tahoma Creek, A. H. Smith and H. Imshaug, July 30 (St. *3779*); Kautz Creek near the Nisqually River, Smith and Imshaug, August 4 (St. *3820, 3829*); lower Tahoma Creek, Smith and Imshaug, August 19 (St. *3925*); lower Nisqually River near Kautz Creek, Smith and Stuntz, August 14 (St. *3958*); Longmire, along the Trail of the Shadows, A. H. Smith, August 21 (St. *4051*); lower Tahoma Creek, Smith and Stuntz, September 8 (St. *4395*), September 12 (St. *4430*), and again, Smith, M. McKenny and Stuntz, September 19 (St. *4619*).

This *Inocybe* was found throughout the collecting season, but never in large numbers: all the collections cited except the one shown in figure 6 consist of but one or two carpophores.

Its rather pale yellowish gray squamulose pileus and fibrillose lilac stipe make it one of the more readily recognizable species of the *Inocybe obscura* group. Our specimens agree well with Lange's description and illustration (**1938**, p. 73, pl. 111–F), except for having a somewhat more robust stature. Heim considers Lange's species to be synonymous with *I. obscura* var. *violascens* (Quél.) Heim, but the spores he illustrates (**1931**, p. 256) for the latter differ enough in size and shape from those given by Lange (**1938**, p. 73, pl. 11–F) for *I. griseo-lilacina*, as well as from those of our own collections, to raise a reasonable doubt as to the advisability of merging the two.

Inocybe laetior Stuntz, sp. nov. *Figs. 5g, h; 7*

Pileus 2–3.5 cm. latus, e campanulatus expansus, umbonatus, sericeus, siccus, glaber, margine interdum subrimosus, centro fulvus vel badius, margine melleus vel olivaceo-brunneus; caro pallida, immutabilis, odor vix notabilis; lamellae adnexae, ventricosae, 6–7 mm. latae, ex pallidis olivaceo-brunneae; stipes 2.5–5 cm. longus, 2.5–5 mm. crassus, aequalis, basis leviter bulbosus vel non, omnino sericeus, subnitens, longitudinaliter striatus, laete salmoneus vel incarnatus, basis albus; sporae 9–11 (13) × 5.5–6.5 μ, leves, amygdaliformes; pleurocystidia (53) 60–80 × 20–30 μ, ventricosa.

Pileus 2–3.5 cm. broad, campanulate and umbonate, becoming expanded and finally plane or nearly so, with a low rounded umbo, the margin incurved at first, then slightly rounded, finally plane and more or less radially split, surface smooth, appressed silky, the cuticle dense, compact, remaining unbroken at the center, becoming more or less rimose toward the margin, usually not at all scaly but sometimes becoming so in dry weather, color at first uniformly

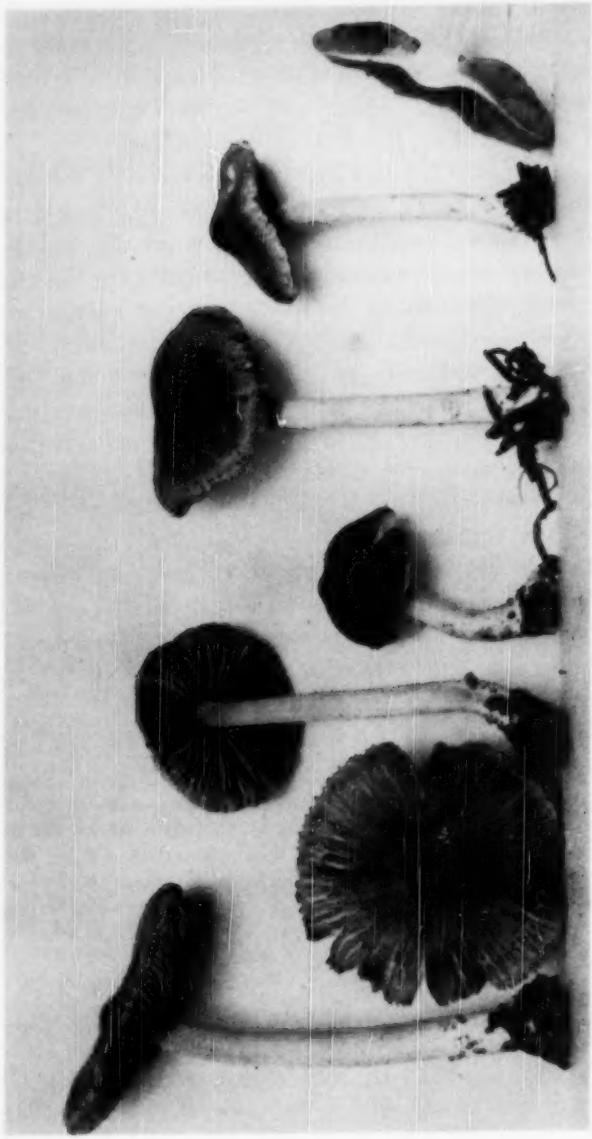

FIG. 7. *Inocybe lacera*, ×1. Photo D. E. Stuntz

brown, then becoming brassy yellowish at the margin and the center remaining tawny brown, center gold brown 14G12, "auburn" or "Sanford's brown," margin at first 14H10 becoming centennial brown 13K8 to Isabella 13K7 or honey 12J6, rarely as dark as "Mars brown" in the center, and shading out through "Verona brown" to "Sayal brown" at the margin, marginal area often more or less virgate with the tawny cuticular fibrils of the center; context 2–3 mm. off the disc, white or pallid, unchanging on exposure, odor slight, spermatic-subraphanoid; lamellae adnexed or sometimes narrowly adnate, often minutely uncinate, broadly rounded at the stipe, bluntly rounded at the margin, asymmetrically ventricose, widest toward the margin, rather broad, 6–7 mm., not close, 30–35 reach the stipe, with about as many inserted, edges slightly serrulate in most specimens, color at first pallid, about sheepskin 11C3 or ecru 11C2, becoming 13I6 to 13J6 (pallid olivaceous), finally khaki 13J7 (dull olivaceous brown); stipe 2.5–7 cm. long, 2.5–5.5 mm. thick, terete or sometimes a little compressed, equal, the base usually slightly bulbous, solid, the context pale salmon-incarnate to yellowish incarnate, unchanging on exposure, surface satiny, somewhat shining, everywhere white-pruinate, finely but notably longitudinally grammate, the base densely white-myceloid, color bright salmon incarnate, often becoming paler with age (auteuil 11C7, 11B8, formosa 12A8, "light pinkish cinnamon," "pinkish cinnamon," or "vinaceous cinnamon," rarely as dark as "cacao brown," Mindoro 13A8 or 11D8), base persistently white.

Spores 9–11 (13.5) × 5.5–6 (6.5) μ, smooth, inequilaterally amygdaliform; pleurocystidia (53) 60–80 × 20–30 μ, ventricose, with very short pedicel and obtuse apex, thick-walled; cheilocystidia mostly like the pleurocystidia in size and shape but a few small clavate thin-walled cells also present.

Under conifers, lower Tahoma Creek, Smith and H. Imshaug (St. *3914*), August 18 (St. *4019*), September 12 (St. *4424*); A. H. Smith, September 12 (St. *4432*), September 12 (St. *4473*), Smith, September 14 (St. *4506*), September 18 (St. *4616*); Smith and Margaret McKenney, Tahoma Camp Grounds, September 20 (St. *4655*—**type**); Smith, lower Tahoma Creek, September 21 (St. *4668*); September 21 (St. *4675*); September 23 (*4710, 4712*).

This species can be recognized quite easily by its strikingly colored salmon-incarnate stipe and its pileus with red-brown center and brassy yellowish margin. These colors appear to be quite constant; of the twelve collections made, only one (*4712*) had

both pileus and stipe somewhat darker than the others. The two species most closely related to *Inocybe laetior* are *I. obscura* var. *rubens* Heim and *I. subdestricta* Kauffman. From both of these *I. laetior* differs in its non-uniformly colored pileus, and especially in its much more brightly colored salmon-incarnate stipe. Its spores are about intermediate in size between those of the other two species, and its pleurocystidia resemble those of *I. obscura* var. *rubens* rather closely.

Photo D. E. Stuntz

FIG. 8. *Inocybe leiocephala*, × 1.

Inocybe leiocephala Stuntz, sp. nov. *Figs.* 5*i*, *j*; 8

Pileus 1.2–3.5 cm. latus, ex obtuse campanulatus expansus, umbonatus, glaber, centro obscure brunneus, margine castaneus vel rubrobrunneus; cutis compacta, hygrophana, nitens; caro firma, alba, immutabilis; odor nullus; lamellae adnatae, postice sinuatae, ventricosae, 4.5–5 mm. latae, ex pallidis olivaceo-brunneae; stipes 1.5–5 cm. longus, 2.5–5.5 mm. crassus, aequalis,

basis rare leviter bulbosus; omnino leviter pruinatus, subnitens, manifeste longitudinaliter striatus, incarnatus vel incarnato-cinnamomeus; sporae 8.5–12 × 5.5–6.5 μ, leves, inaequales; pleurocystidia 56–73 × 15–25 μ, ventricosa, muro valde crasso praedita.

Pileus 1.2–3.5 cm. broad, obtusely campanulate becoming expanded, with a low broad umbo and persistently rounded margin, surface smooth, glabrous, innately silky and shining, the cuticular layer dense, compact, rather hygrophanous, remaining unbroken or occasionally a little diffracted in places when dry, color "warm sepia" to "mummy brown" on the umbo, elsewhere varying from red-brown to dingy brassy yellow-brown, usually "russet," "auburn," "Sanford's brown," margin sometimes nearer "buckthorn brown," or sometimes the whole pileus uniformly "chestnut brown"; context 1–2 mm. off the disc, hard, white, unchanging on exposure, odor none, or very faintly spermatic; lamellae narrowly to broadly adnate, rounded-sinuate to emarginate, bluntly pointed at the margin, subventricose, 4.5–5 mm. broad, moderately close, color at first pallid, becoming olivaceous brown (Isabella 13K7, 14F7, syrup 14L8, finally darker, 15J10); stipe 1.5–5 cm. long, 2.5–5.5 mm. thick, terete or occasionally somewhat compressed, equal, base not bulbous, or rarely very obscurely so, solid, context pallid with a flush of incarnate, unchanging on exposure, surface satiny, shining, conspicuously longitudinally hygrophanous-grammate, pruinate all over, color incarnate, "pinkish buff" to "light pinkish cinnamon," becoming "pinkish cinnamon" to "cinnamon" (papyrus 12C7 or 13E7), finally "Sayal brown" in age.

Spores 8.5–10 (12) × 5.5–6.5 μ, somewhat variable in size, inequilateral; pleurocystidia 56–73 × 15–25 μ, ventricose with long neck and short pedicel, very thick-walled; cheilocystidia of two kinds, one the same size and shape as the pleurocystidia, the other thin-walled, clavate, 22–34 × 8–13.5 μ.

Habit, habitat and distribution: Under conifers, junction of Fish Creek with Tahoma Creek, Smith, Knowles and Stuntz, August 9 (St. *3929*); camp grounds, Longmire, Smith, August 31 (St. *4249*); in moss under Douglas fir near the inn, Longmire, September 1 (St. *4259*); same locality, September 24 (St. *4739*—**type**), and September 27 (St. *4816*).

Discussion: The very compact layer of cuticular hyphae gives the pileus surface of this species a glabrous, hygrophanous appearance unusual in *Inocybe*, and very similar to that of many of the small brown Cortinarii of the subgenus Hydrocybe. This characteristic, the rather dark red-brown pileus, small compact stature,

incarnate stipe, and thick-walled pleurocystidia, are the principal distinguishing features. While the pileus usually has a very dark brown umbo and red-brown margin, it may be uniformly bay in color, or even almost a mahogany red, and is apt to become somewhat paler and more yellowish brown on drying. The most closely related species appear to be *Inocybe subdestricta* Kauffman, *I. obscura* var. *rubens* Heim, and *I. laetior* Stuntz, in diminishing order of resemblance. From all three of these *I. leiocephala* differs markedly in the nature of its pileus surface: it has smaller spores than *I. obscura* var. *rubens*, and differs further from *I. laetior* in the color of both pileus and stipe.

INOCYBE OBLECTABILIS Britzlmayr, fma. DECEMGIBBOSA Kühner
Fig. 9; 10*a*, *b*

Pileus 2–4 cm. broad, campanulate or conic-campanulate, expanding and becoming shallowly campanulate and finally almost plane, with a broad rounded umbo, margin incurved, usually becoming almost plane, often undulating and more or less broadly lobed, surface at first with a thin white webby coating of surface fibrils which persist for a time at the center as pale webby patches, finally vanishing entirely, cuticle silky smooth, lubricous when moist, shining when dry, remaining unbroken at the center, becoming conspicuously long-rimose toward the margin, color "Sayal brown" to "cinnamon brown," "Verona brown," 14G9, or burnt umber 15A12, the umbo often considerably paler (raffia 11E5 or pallid ivory); context 2–2.5 mm. thick off the disc, firm, white, unchanging on exposure, odorless; lamellae adnexed and uncinate, shallowly to deeply rounded-emarginate, pointed at the margin, oblong-subventricose, 4–6.5 mm. broad, moderately close, color at first pallid gray (putty 11B2), becoming darker and more olivaceous (grain 13B5 to cracker 13D6, finally 14I7 to maple sugar 14J8); stipe 2.5–7 cm. long, 3–7 mm. thick, terete or compressed, equal, the base with a conspicuous flat marginate bulb 7–13 mm. in diameter, solid, the context pallid or yellowish, usually with a flush of incarnate at the surface, unchanging on exposure, surface satiny, more or less shining, everywhere finely white-pruinate and conspicuously longitudinally hygrophanous-grammate, color pale yellow, usually with more or less of a flush of incarnate (10C5, or "light ochraceous buff," "pinkish buff," "light pinkish cinnamon," or "pinkish cinnamon"), the base persistently white.

Spores 9–11 × 6.5–8 μ, oblong, with an average of eight rather coarse nodules; pleurocystidia 45–67.5 × 13.5–20 μ, fusiform-

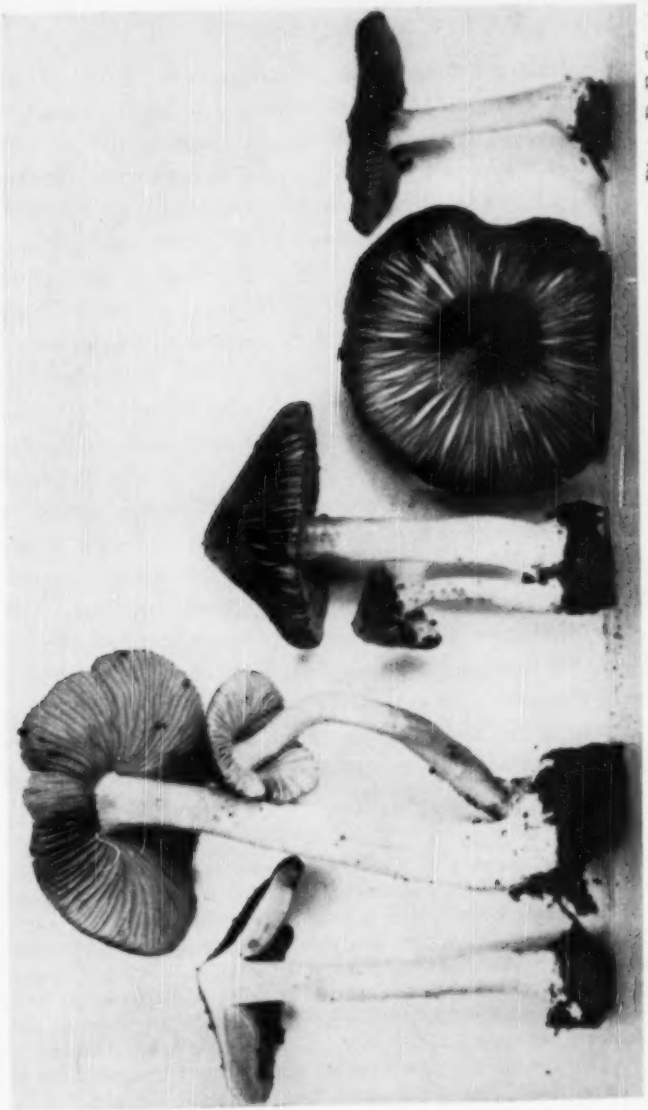

FIG. 9. *Inocybe oblectabilis* f. *decemgibbosa*. × 1. Photo D. E. Stuntz

ventricose with short neck and pedicel, moderately thick-walled; cheilocystidia like the pleurocystidia in size and shape, also thin-walled and clavate, 22.5–30 × 9–11.5 μ.

Habit, habitat and distribution: In a lawn under conifers, Longmire, August 4 (St. *3811*); under conifers, lower Tahoma Creek near the highway, Knowles and Stuntz, August 16 (St. *3980, 3981*); lower Tahoma Creek at the old campgrounds, Smith & Stuntz, August 20 (St. *4043*), same locality, August 22 (St. *4097*), September 8 (St. *4388*), September 12 (St. *4459*), and September 18 (St. *4612*); lower Tahoma Creek, conifer woods one half mile from the highway, September 21 (St. *4674*), same locality, Smith, September 21 (St. *4680*) and again September 23 (St. *4706*).

Discussion: Neither *Inocybe oblectabilis* Britz. nor its two forms have been reported previously from North America. The form *decemgibbosa*, however, is apparently not uncommon in Washington, having been collected by the junior author in several localities outside the Park. The incarnate color of its stipe is apt to vary from a slight flush to a decidedly salmon pink cast with different collections, and may even be lacking at the time the specimens are gathered, only to develop later after they have stood for a while. The other distinctive features (smooth, brown, rimose pileus and pallid grayish lamellae) are quite constant, however, and the ensemble of characteristics makes this an easily recognized *Inocybe*. The brown pileus and incarnate stipe readily separate it from *Inocybe praetervisa* Quél. and related species.

Inocybe picrosma Stuntz, sp. nov. *Figs.* 10*c, d;* 11

Pileus 1.5–4 cm. latus, e campanulato expansus et late umbonatus, siccus, primo sericeo-fibrillosus, postremo plus minusve squamuloso-diffractus, cremeus vel pallide luteus; caro pallida, immutabilis, odor valde specialis, aliquatenus raphaneus et resinosus, etiam acer; lamellae rotundato-adnexae vel anguste adnatae, ventricosae, 3.5–5 mm. latae, ex pallidis griseo-olivaceae; stipes 2.5–8.5 cm. longus, 3–7 mm. crassus, aequalis, basis plerumque abrupte bulbosus sed non marginatus, primo omnino albo-pruinatus, deorsum glabrescens, longitudinaliter striatus, pallide luteolus, aetate sordide brunneus vel avellaneus; sporae 8.5–10 × 5 μ, leves, amygdaliformes; pleurocystidia 40–60 × 16.5 μ, subclavata.

Pileus 1.5–4 cm. broad, campanulate and umbonate, becoming campanulate-expanded, the margin at first narrowly involute, long remaining broadly rounded, surface appressedly silky-fibril-

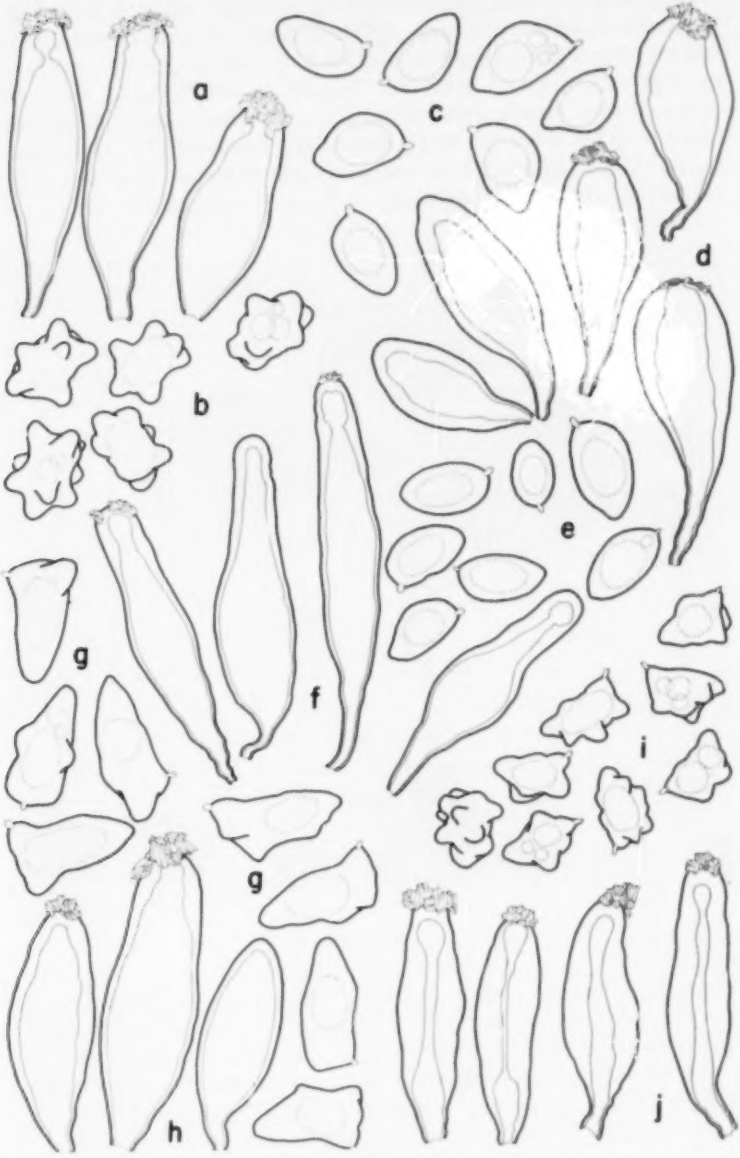

FIG. 10. Spores and cystidia of *Inocybe*.

lose, smooth, shining, some remaining so at all times, but the majority becoming more or less diffracted-scaly at least at the center and usually clear to the margin, color uniformly pale creamy yellow to pale buff, "cream buff," to "Naples yellow," 10F4, 11F5, or chamois 1115, sometimes more or less flushed with "cinnamon buff" to "clay color" or even darker (macaroon 12H7) especially on the disc; context 1.5–3 mm. thick off the disc, firm but quite brittle, pallid to pallid yellowish, unchanging on exposure, odor very

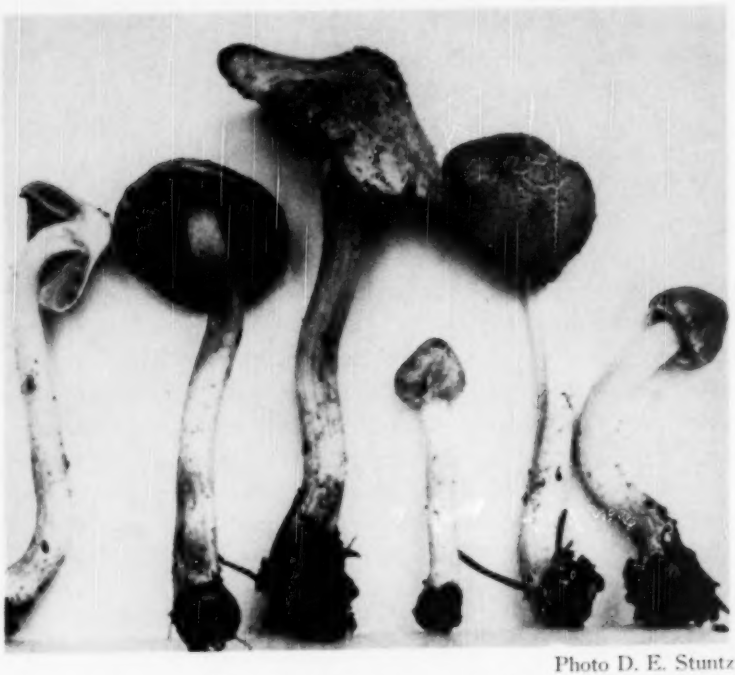

Photo D. E. Stuntz

FIG. 11. *Inocybe picrosma*, × 1.

characteristic, not strong, but very penetrating, spermatic for an instant when the context is first exposed, but immediately becoming quite complex, predominantly a mixture of raphanoid and resinous with a trace of acetic acid, having a very decided pungency which quickly produces a tingling sensation in the back of the throat; taste a little astringent and acrid at first, soon becoming mild and almost sweet, but leaving an unpleasant aftertaste; lamellae adnexed to narrowly adnate, abruptly and deeply rounded at the stipe, broadly rounded at the margin, ventricose, moderately broad,

4–5 mm., moderately close, about 90 reaching the stipe, as many more inserted, color at first pallid (parchment 12B3, cream 9D2, "ivory yellow"), becoming rather pale grayish olivaceous brown (tanaura 12D4, 12F5, prairie 13F6); stipe 2.5–8.5 cm. long, 3–7 mm. thick, terete, equal, the base almost always with a narrow flat bulb which in some specimens may be emarginate, solid, context white or pallid, unchanging on exposure, but slowly in age becoming suffused all over with "avellaneous" or dull vinaceous brown or fuscous brown, surface satiny, shining, conspicuously longitudinally hygrophanous-grammate, entirely white-pruinate at first, remaining so at apex, becoming almost glabrous below, color very pale yellow ("cartridge buff," "ivory yellow," 9D1), in age becoming flushed first at the apex then all over with dull vinaceous brown ("avellaneous," "wood brown," cork 12B7, India spice 13B8) or fuscous brown ("drab," "cinnamon drab," winter leaf 15A8), the margin of the bulb almost always with a flush of "apricot buff," "ochraceous salmon" or "light ochraceous salmon."

Spores 7–8.5–10 (11) × 5–6 μ, smooth, inequilaterally amygdaliform; pleurocystidia 36–50 (63) × 16.5–21 μ, fusiform-clavate, with obtuse apex and slender pedicel, very thick-walled above; cheilocystidia of two kinds, some similar to the pleurocystidia in size and shape, others clavate, thin-walled, 22–56 × 8–11.5 μ.

Habit, habitat and distribution: Under conifers, lower Tahoma Creek, Smith, September 14 (St. *4536*); same locality, September 18 (St. *4647*); Tahoma Creek Camp Grounds, Smith, September 20 (St. *4660*); lower Tahoma Creek, Smith and Stuntz, September 21 (St. *4700*); same locality, September 23 (St. *4736*); lower slopes of Tumtum Mountain, near Tahoma Creek, Smith and Stuntz (St. *4839*—**type**).

Discussion: This *Inocybe* can be recognized quite readily by its silky pale yellow pileus, pallid yellow stipe which turns dull vinaceous brown in age, and especially by its unique odor. In coloration and general appearance it resembles *Inocybe Kauffmanii* Smith, and indeed the two species appear to be rather closely related; however, the stipe of *I. Kauffmanii* does not become vinaceous brown with age, and its odor is entirely different. There is also a superficial resemblance between *I. picrosma* and *I. suaveolens* Stuntz, but no real relationship whatever, as the two have entirely different microscopic characteristics as well as very different odors. The odor of *I. picrosma* is its chief distinguishing

characteristic; though not actually strong, it has a peculiar pungency that sets it apart from most fungus odors and usually produces an effect similar to slight traces of sulphur dioxide. The brittleness and darkening with age characteristic of the stipe context are well marked, and seemed constant in the various collections. The stipe context is so brittle that it is difficult to collect a specimen without breaking off either the base or the pileus. The stipe of older specimens is apt to become gray in drying, like that of *Inocybe xanthomelas* Kühner.

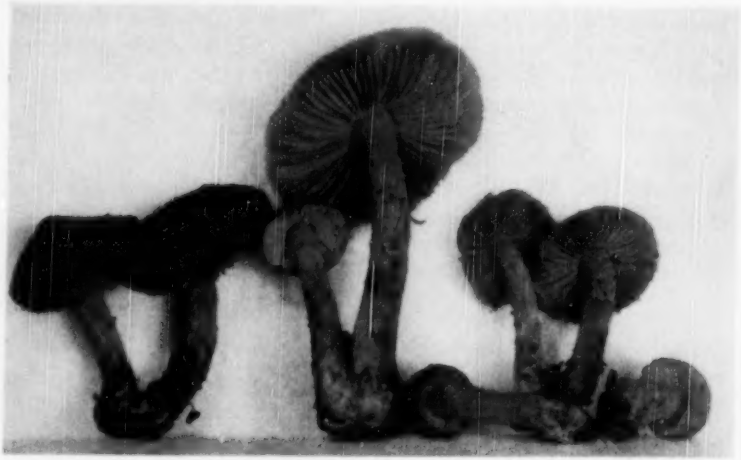

Photo D. E. Stuntz

FIG. 12. *Inocybe pyrotricha.*

Inocybe pyrotricha Stuntz, sp. nov. *Figs.* 10*e, f;* 12

Pileus 1.5–3 cm. latus, campanulatus demum expansus, late umbonatus, siccus, mox squamulosus, centro ferrugineus vel rufus, margine pallescens, sordide brunneus; caro pallida, immutabilis, odor tenuiter raphaninus; lamellae adnatae, ventricosae, 6 mm. latae, primo violaceae demum pallide brunneae; stipes 2.5–5 cm. longus, 2.5–5 mm. crassus, aequalis, solidus, sursum subglaber, pallide violaceus, deorsum fibrillis rufis dense obtectus; sporae 7–10 × 4.5–5 μ, leves, amygdaliformes; pleurocystidia 66–80 × 16.5 μ, ventricosa, pedicellata.

Pileus 1.5–3 cm. broad, campanulate, becoming campanulate-expanded with a large broadly rounded umbo, the margin entire, at first incurved, becoming gently rounded, finally almost plane; surface dry, densely appressed-fibrillose, smooth only in the youngest buds, soon diffracted scaly with small flat frequently re-

curved squamules at least in the center, squamulose almost to the margin in some, in others the marginal area merely fibrillose-lacerate and more or less rimose, decorated at first with the remains of the avellaneous cortina, soon denuded, color of young buds uniformly ferruginous or rufous, of older pilei "burnt sienna" to "Kaiser brown," "ferruginous" or "cinnamon rufous" at the center, some shade of dingy brown such as cork 12B7, 13E9, Mosul 14F8 toward the margin, on drying becoming generally somewhat paler, "ochraceous tawny" to "tawny" at the center, toast 13F8 at the margin, the surface fibrils finally assuming a decidedly yellow-orange cast as they dry out; context thin, firm, at first with a pale lavender tinge, soon pallid, unchanging on exposure; odor rather pleasant, subraphanoid or faintly spermatic, taste mild, a little nutty; lamellae adnate, deeply and broadly rounded at stipe and margin, symmetrically ventricose, rather broad, up to 6 mm., moderately close, about 50 reaching the stipe, with another series inserted in various lengths, and occasional marginal lamellae, color at first "pale bluish lavender" to "pale verbena violet" or "lavender" becoming "light cinnamon drab," finally adobe 14D7; stipe 2.5–5 cm. long, 2.5–5 mm. thick, terete, equal, the base a little clavate-incrassated and often abruptly truncate, solid, the context pallid within, yellow at the surface toward the base, persistently pale lavender or bluish lavender at the surface toward the apex, unchanging on exposure, surface densely peronate with a floccose-fibrillose sheath which persists on the basal two thirds, there becoming pulled, as the stipe expands, into conspicuous longitudinal streaks and reticulations which diminish in abundance upwards, the apex subglabrous, finely pruinate and minutely fibrillose, ground color at the base "Naples yellow" to "Colonial buff," at the apex "pale bluish lavender" to "lavender," the median part pallid, color of the peronate surface fibrils "Sanford's brown" to "burnt sienna" or "orange rufous" at the base, shading upward through "cinnamon rufous" to "avellaneous" near the apex, in young specimens the base is usually "zinc orange" to "ochraceous orange."

Spores 7–10 × 4–5–6 μ, usually 8–9 × 5.5 μ, with broadly rounded ventral profile; pleurocystidia 66–80 × 13.5–16.5 μ, slender, ventricose with long neck and well-defined pedicel, thick-walled; cheilocystidia of two kinds, one similar in size and shape to the pleurocystidia, the other thin-walled, clavate, 20–38 × 11–26 μ, in dense clusters, both kinds with pale brown wall.

Habit, habitat and distribution: Under conifers, base of Rampart Ridge near Longmire, September 15, collected by Smith (St. *4545* —**type**).

Photo D. E. Stuntz

FIG. 13. *Inocybe rainierensis*, × 1.

Discussion: This is certainly one of the most brightly colored and distinctive of the group of species closely related to *Inocybe obscura* (Fr.) Gill. The coating of rusty red fibrils on the stipe and the decidedly reddish color of the pileus surface are the features which readily distinguish it from the other members of its stirps.

Inocybe rainierensis Stuntz, sp. nov. *Figs.* 10*g, h;* 13

Pileus 1.5–4.5 cm. latus, obtuse campanulatus demum expansus, late umbonatus vel non, siccus, sericeus, centro velamine pallide brunneo obtectus, cutis obscure brunneo; caro firma, pallida, immutabilis; odor nullus vel vix raphaneus; lamellae adnatae, subconfertae, ventricosae, 4.5–7 mm. latae, ex pallidis brunneo-olivaceae; stipes 2–4.5 cm. longus, 3.5–8 mm. crassus, plerumque compressus, aequalis, basis bulbosus, sericeo-nitens, longitudinaliter fibrilloso-striatus, sursum cinnamomeo-incarnatus, deosorum brunneus; sporae 10–15 × 5–6.5 μ, crasse 4–5-tuberculatae, apice praelongae; pleurocystidia 60–86 × 16–20 μ, fusoideo-ventricosa.

Pileus 1.5–4.5 cm. broad, obtusely campanulate, becoming expanded and obtusely convex to broadly umbonate, margin persistently broadly rounded; surface dry, smooth, covered at the center with a thin, webby, appressed, persistent pallid brown coat of fibrils, true cuticle appressed-fibrillose, silky smooth, finally becoming more or less obscurely areolate to subscaly at the center, margin remaining smooth or in some finally a little rimulose, usually decorated with the rather copious pallid veil, color dark brown with an obscure reddish cast ("Verona brown," "snuff brown," Montella 8J11, 8J12, or "bister" to café noir 8H12 at center, elsewhere brownstone 7C10, Vandyk brown 7A11, dark beaver 15A9, teakwood 15C9, English oak 15A10), appearing more pallid brown (adobe 14D7) here and there because of the coating of surface fibrils; context 2–3 mm. thick off the disc, firm, pallid or tinged with brown, unchanging on exposure, odor none or faintly raphanoid; lamellae narrowly to broadly adnate, sinuate to emarginate, more or less deeply rounded at the stipe, bluntly pointed at the margin, ventricose, 4.5–7 mm. broad, moderately close, 55–65 reaching the stipe, the same number inserted in irregular lengths, color pallid at first (putty 11B2, becoming Malacca 12C4), finally olivaceous brown (airdale 14F6, 14G6, 14G7); stipe 2–4.5 cm. long, 3.5–8 mm. thick, terete or more often markedly compressed, equal above the base which has an abrupt to napiform bulb 7–11 mm. in diameter, solid, unchanging on exposure, surface densely longitudinally silky fibrillose at first, becoming longitudinally fibrillose-streaked and hygrophanous-grammate on a satiny shining surface, apex pruinate, margin of bulb densely beset with thick

matted fibrillose tufts of the copious veil, color at first uniformly "light pinkish cinnamon," darkening and finally becoming brown below, at length with the apex pallid, shading down through suntan 13B7, Tuscan tan 13C8, marron glace 14A8, and finally "Verona brown," with the base eventually "warm sepia," but all these colors appearing paler because of the pallid surface fibrils.

Spores (10) 11.5–13 (16) × 5.5–9 μ, quite irregular in outline, but generally with 4 or 5 coarse nodules at the base, and the apex prolonged into a bullet-shaped structure; pleurocystidia 58–70 (78) × (12) 16–25 μ fusiform-ventricose, only moderately thick-walled; cheilocystidia same size and shape as the pleurocystidia, accompanied by a few small and inconspicuous clavate ones.

Habit, habitat and distribution: Under conifers, elevation 4800 ft., Eagle Peak, Smith, August 26 (St. *4161*); under Abies, Reflection Lake, Smith, August 28 (St. *4200*—**type**).

Discussion: This *Inocybe* and *Inocybe chelanensis* Stuntz have practically the same microscopic characters, but differ markedly in outward appearance. The pileus of *I. rainierensis* is much darker and more uniformly colored than that of *I. chelanensis*, and the stipe always has a conspicuous bulb, whereas the stipe of *I. chelanensis* lacks a bulb altogether. Unfortunately, *I. chelanensis* seems to be a rare species, as yet known only from the type collection, so that nothing can be said concerning the extent of its variability with respect to the above characteristics. In view of this situation, and since there is actually such a marked difference in the appearance of the two fungi, it seems best to consider them separate though closely related species.

Inocybe suaveolens Stuntz, sp. nov. *Figs.* 10*i*, *j*; 14

Pileus 2–4.5 cm. latus, campanulato-expansus, siccus, squamosus, centro albidus, margine primo albidus deinde pallide luteus vel cremeus; caro alba vel pallida, immutabilis, odor fragrans; lamellae adnexae, ventricosae, 4–5 mm. latae, ex albidae brunneo-olivaceae; stipes 3.5–8.5 cm. longus, 2.5–7 mm. crassus, aequalis, basis marginato-bulbosus, solidus, caro pallide luteola, immutabilis, longitudinaliter striatus, omnino albido-pruinatus, pallide luteus, tarde sordide brunneus vel subincarnatus; sporae 7–9 (10) ×5.5–6.5–7.5 μ, nodulosae vel subnodulosae; pleurocystidia 43–58 (60) × 10–16.5 μ, fusoidea vel sublanceolata, muro valde crasso praedita.

Pileus 2–4.5 cm. broad, campanulate, becoming expanded and broadly umbonate, margin involute then gently rounded, finally almost plane, surface dry, silky-smooth and subshining at first

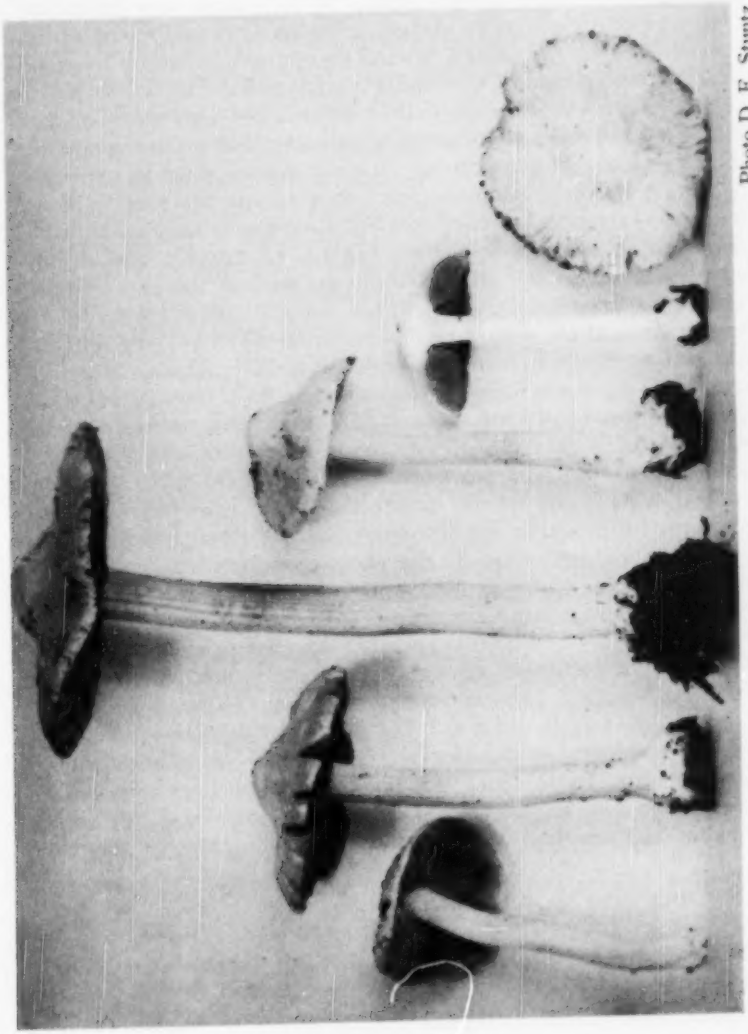

Photo D. E. Stuntz

FIG. 14. *Inocybe sarcolema*, ×1.

(lubricous when thoroughly wet), soon becoming more or less diffracted scaly, the scales with free ends, or even more or less conspicuously revolute, margin often rimose, sometimes becoming excoriate or deeply split in age, color at first uniformly white, remaining so at the center, toward the margin becoming more or less yellowish or tinged with buff ("cartridge buff" to "cream buff," or maple 11E4 or chamois 11I5); context 2–3 mm. thick off the disc, hard, white or pallid, unchanging on exposure, odor spermatic for an instant when the context is first cut open, then immediately becoming and long remaining very fragrant, almost exactly like a mixture of the odors of sweet pea and *Convallaria majalis;* lamellae adnexed, in some almost free, slightly to broadly and deeply rounded close to the stipe, bluntly pointed at the cap margin, ventricose, moderately broad (4–5 mm.), rather close, 60–70 reaching the stipe, with 2 or 3 series inserted in varying lengths, color at first pallid, becoming India buff 12E5, finally brownish olivaceous (Isabella 13K7 to maple sugar 14J8, bronze 14L9, or 14G7); stipe 3.5–8.5 cm. long, 2.5–7 mm. thick, terete or somewhat compressed, frequently flexuous, base with a distinct and sometimes conspicuous marginate bulb 6–12 mm. thick, solid, the context hard, often spirally twisted, white to pale yellowish, unchanging on exposure but becoming a little flushed with brownish or incarnate at the surface in age, surface somewhat shining, everywhere densely white-pruinate, conspicuously and broadly longitudinally hygrophanous-grammate, color at first "cartridge buff" becoming "ivory yellow" to chamois 11I5, then in age more or less flushed with brownish or incarnate shades, such as "cinnamon buff," Pablo 12G7, or even Centennial brown 13K8.

Spores 7–9 (10) × 5.5–6.5 (7) μ, merely angular to decidedly nodulose; pleurocystidia 43–58 (60) × 10–16.5 μ, subfusiform to sublanceolate, apedicellate, very thick-walled; cheilocystidia of two kinds, one like the pleurocystidia in size and shape, the other smaller, clavate, thin-walled, 15–22.5 × 9–11 μ, in dense clusters.

Habit, habitat and distribution: Under conifers, lower Tahoma Creek, Smith and Stuntz, September 18 (St. *4646*—**type**); lower slopes of Tumtum Mountain, along lower Tahoma Creek, September 26 (St. *4800*).

Discussion: The white silky pileus which becomes scaly and more or less flushed with yellow, the pruinate, marginate-bulbed, pale yellow stipe, and especially the persistent, sweet odor, closely resembling that of the common sweet pea, are the distinguishing characteristics of this species. There is no tendency of any part

of the pileus or stipe context to become reddish on exposure to air, as is usually the case with Inocybes having an aromatic odor. The most closely related species appear to be *Inocybe Bresadolae* Massee sensu Kühner (**1932**, p. 158) and *I. capucina* Fr. sensu Patouillard as described by Heim (**1931**, p. 290). From both of these, *I. suaveolens* differs in the much paler color of its pileus, and the complete lack of any change to red in the exposed context. To judge from the descriptions cited above, the odor of *I. suaveolens* is also different, but probably not too much emphasis should be placed on a characteristic as difficult to judge objectively as odor.

Mycena fallax Smith sp. nov. *Fig. 3e, f*

Pileus 2–5 mm. latus, obtusus demum convexus vel planus, undulatus et plicato-striatus, glaber, udus, aquose albidus deinde candidus, membranaceus; lamellae angustae, subdistantes, praedecurrentes, albidae; stipes 0.5–1 cm. longus, filiformis, glaber, albidus, insiticius; sporae 7–8.4 × 3–4 μ anguste ellipsoideae, amyloideae; pleurocystidia fusoide ventricosa, apicibus echinulatis, 40–60 × 9–13 μ; cheilocystidia pleurocystidia similis.

Pileus 2–5 mm. broad, obtuse becoming convex, expanding to broadly convex or plane, the margin connivent to stipe at first, surface undulating and plicate-striate, glabrous and moist, watery-white or shining-white over margin and watery-white on disc, shining white over all when faded; flesh membranous and fragile; lamellae narrow, subdistant to distant, long-decurrent on the enlarged apex of the stipe, white like pileus, edges even; stipe 0.5–1 cm. long, filiform or about 0.25 mm. thick, hyaline-white and perfectly naked (no pruinosity on youngest buttons), base inserted on the substratum.

Spores 7–8.4 × 3–4 μ, narrowly ellipsoid, smooth, distinctly amyloid when fresh, weakly amyloid after standing in herbarium two years; basidia four-spored, 26–30 × 7–8 μ; pleurocystidia 40–60 × 9–13 μ, fusoid-ventricose, the apices simple to forked and with scattered echinulations, thin-walled and hyaline in KOH; cheilocystidia similar to pleurocystidia; cuticle of pileus of narrow filaments 2–3.5 μ in diam. and sparsely covered with short rod-like projections; caulocystidia present only near the gills and similar to pleurocystidia.

Habit, habitat and distribution: Gregarious on *Rubus* canes in very wet localities, Sandy, Oregon, November 5, 1947 (Sm. *28505* —**type**). Two fruiting bodies were found in the Park in a dense

growth of *Rubus* near a beaver pond along lower Tahoma Creek late in September, 1948, but the material was used up in the process of identification.

Discussion: This is an easily recognized *Mycena* when fresh because of the echinulate cystidia and because the spores give a stronger amyloid reaction at that time than after the specimens have been dried. The cystidia have coarser echinulations than do those of *M. borealis* Smith. *M. fallax* seems to resemble *M. litoralis* Smith in many characters but has more strongly decurrent gills and echinulate cystidia.

Photo A. H. Smith

FIG. 15. *Mycena rainierensis*, × 1.

Mycena rainierensis Smith sp. nov. *Figs. 3b, d;* 15

Pileus 15–30 mm. latus, convexus demum late convexus, glaber, lubricus, striatulatus, margine albidis, disco pallide griseis; lamellae albidae, latae, late adnatae, confertae vel subdistantes; stipes 5–10 cm. longus, 1.5–3 mm. crassus, aequalis, glaber, albidus; sporae 8–10 μ, punctatae; cheilocystidia 60–120 × 9–14 μ, fusoide ventricosa, apicibus acutis.

Pileus 15–30 mm. broad, convex with a straight to connivent margin, broadly convex in age, surface lubricous when wet, with fine translucent striations nearly to disc, white with a watery gray more or less sharply defined discal spot, some merely watery white to watery grayish over disc; flesh thin, pliant, odor and taste not distinctive; lamellae white, broad, bluntly adnate and horizontal, only moderately close, slightly avellaneous in age when water-soaked, edges even and concolorous; stipe 5–10 cm. long, 1.5–3 mm. diam., equal, glabrous, naked, dull white but shining when wet, transversely translucent-striate, with a slight pruinosity at line of gill attachment.

Spores globose to subglobose, hyaline, 8–10 μ in diam., amyloid, with a very thin amyloid exospore which soon disappears, endospore thicker and perforated with innumerable pores, the pore walls usually slightly amyloid; basidia two-spored and four-spored, 27–32 × 8–9 μ; pleurocystidia none; cheilocystidia 60–120 × 9–14 μ, more or less fusoid-ventricose with subacute to obtuse apices, walls thin and often flexuous, hyaline; gill trama parallel or nearly so, the cells 10–15 μ in diam., cylindric and straight or curved; pileus trama homogeneous, the surface layer of hyphae slightly gelatinous.

Habit, habitat and distribution: Gregarious in a wet area along a stream, Longmire Camp Ground, September 27 (Sm. *31845*—**type**).

Discussion: Only the one collection was found. This species is obviously closely related to *M. bisphaerigera* (Lange) Smith by its spores and gill characters, but differs sharply in having the greatly elongated cheilocystidia as well as paler color. The latter character alone, however, would not be regarded as very significant. *Mycena cineraria* Smith differs in having ellipsoid spores, dark colors, and pleurocystidia. Both the pleuro- and cheilocystidia of *M. cineraria* differ in shape from the cheilocystidia of *M. rainierensis*.

Favre (**1948**) has published a very interesting account of the variants of *Mycena* (*Fayodia*) *bisphaerigera*. He describes

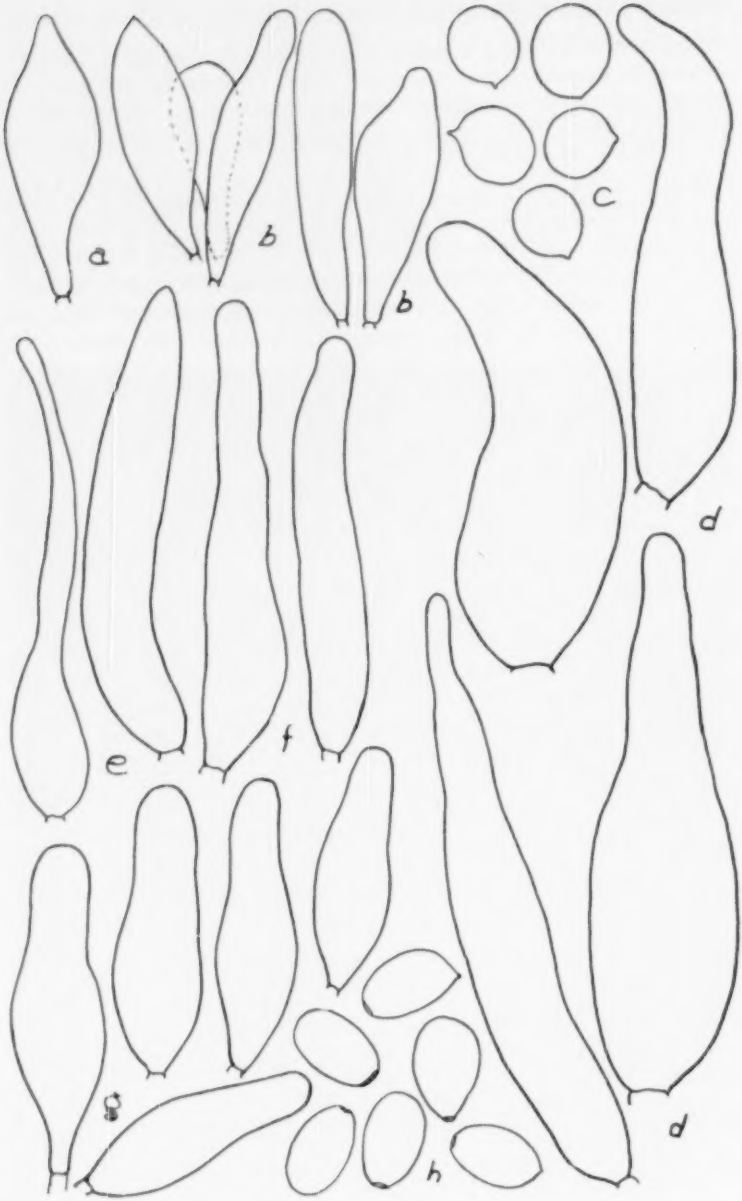

FIG. 16. Spores and cystidia of *Omphalina* and *Psathyrella*.

Fayodia bisphaerigera var. *anthracobia* as a new variety having cheilocystidia like those of *M. rainierensis*, but with the cap very dark in color and the stipe very short (18 × 1.5 mm.). Its spores measure 6–8.5 μ in diam. His variety *longicystis* has more the stature of *rainierensis* but differs in the greatly elongated cystidia. Without question Favre's two varieties and *M. rainierensis* should all be treated as of equal rank. I prefer to regard them as species, and hence describe *M. rainierensis* at that level. This procedure more closely maintains the established concepts of species in *Mycena*, and is certainly of practical value if *Fayodia* is recognized as a genus. As Favre himself pointed out, his varieties differ from each other more sharply than do many related species in other genera, such as *Clitocybe*.

Omphalina isabellina sp. nov. *Fig.* 16*c, d, e, f*

Pileus 5–15 mm. latus, convexus, demum late convexus, impolitus demum subfurfuraceus, hygrophanus, olivaceus vel sub-olivaceus deinde Isabellinus vel subochraceus, striatus; lamellae decurrentes, distantes, latae, olivaceae vel subolivaceae; stipes 1.5–3 cm. longus, 1–2 mm. crassus, aequalis, subolivaceus vel olivaceo-luteus, pubescens demum glaber vel impolitus; sporae 5–6.5 (7) μ, globosae, amyloideae; pleuro- et cheilocystidia nulla; pilo- et caulocystidia ventricosa vel subcylindrica, distinctissima.

Pileus 5–15 mm. broad, convex with a curved in margin when young, broadly convex to nearly plane in age, the margin finally wavy or recurved, surface moist and hygrophanous but with a velvety sheen or minutely scurfy from projecting pilocystidia, "sepia" to dark "Isabella color" or more olivaceous at first, becoming "Isabella color" to "light brownish olive," finally "olive ocher" to "honey yellow" with "light brownish olive" to "Isabella color" striations, sometimes eventually pale yellow and striations inconspicuous; flesh dark olivaceous, pliant but in age fragile, odor and taste not distinctive; lamellae decurrent, distant, broad, tapered to either margin, "deep olive buff" to pallid olivaceous yellow, usually not as yellow as cap and stipe, edges even; stipe 1.5–3 cm. long, 1–2 mm. thick, equal, concolor with cap margin to ocher yellow, more sordid downward, pubescent at first from projecting caulocystidia but in age glabrous to unpolished.

Spores globose to subglobose, 5–6.5 (7) μ, smooth, amyloid; basidia 40–50 × 9–10 μ, four-spored, hyaline in KOH; pleurocystidia and cheilocystidia none; gill trama interwoven, central core of very large cells which are pale yellow in water mounts of

fresh material, merely pale yellow revived in chloral-hydrate-iodine solution and hyaline revived in KOH; pileus trama loosely interwoven, cuticle composed of a turflike covering of large pilocystidia with smoky yellow contents (H_2O mounts of fresh material), the pilocystidia arising from enlarged cells also having smoky yellow contents (when revived in KOH contents of both types of cells dull brown); pilocystidia more or less fusoid ventricose with obtuse apices, 40–70 × 9–16 μ; caulocystidia similar to pilocystidia or with greatly elongated necks above ventricose base, some subcylindric, 40–120 × 7–12 μ.

Habit, habitat and distribution: Gregarious to scattered on very old mossy Douglas fir logs, lower Tahoma Creek, August 14, Stuntz (Sm. *30087*—**type**). A previous collection of the same species was made by the senior author on a similar Douglas fir log at Rhododendron, Oregon, October 14, 1944 (Sm. *19695*).

Discussion: This species is very easily recognized by the Isabella to olive colors, velvety cap and hoary to pubescent stipe. It is most interesting, however, in view of recent attempts to revise generic concepts among the white spored agarics. This species is excluded from *Mycena* in the concept of Smith (**1947**) by having decurrent gills and an incurved cap margin. Singer (**1942**, pp. 128–9) published a short key separating the Marasmioideae with amyloid spores. *O. isabellina* has some of the characters of his *Hydropus*, but does not fit readily into any of the genera. Actually, our species does not appear to belong in Singer's Marasmioideae in spite of a faint resemblance in stature and consistency to *Xeromphalina campanella*. *O. isabellina* does, of course, fall readily into the Friesian genus *Omphalia* (*Omphalina* if the International Rules are followed), but would doubtless be excluded by those who restrict *Omphalina* to species with nonamyloid spores. However, we prefer to place it here at least for the present, where it can be found easily.

Psathyrella alboalutacea Smith sp. nov. *Figs. 3g, h; 16a, b; 17*

Pileus (2) 3–5 (6) cm. latus, obtusus vel convexus, demum planus vel subumbonatus, siccus, innato-fibrillosus demum squamulosus, squamulis albidis demum alutaciis; margo appendiculatus; lamellae adnatae, secedentes, confertae, angustae, latae, albidae demum subfuscae ("hair brown"); stipes 3–5 cm. longus, 10–12 mm. crassus, durus, farctus, intus albidus vel deorsum incarnatus, deorsum fibrillosus, sursum striatus; sporae 7.5–9 × 4–4.5 μ;

pleurocystidia clavata vel subvesiculosa, pedicellata, 25–40 × 10–14 μ, vel fusoidea et apicibus obtusis vel subacutis, 40–53 × 9–14 μ.

Pileus (2) 3–5 (6) cm. broad, obtuse to convex at first, becoming plane or nearly so, the disc in some elevated as a low broad umbo, surface dry and fibrillose, the fibrils innate and forming an

Photo A. H. Smith

FIG. 17. *Psathyrella alboalutacea*, × 1.

appressed, cottony mat over the buttons which are pure white, the fibrils soon becoming aggregated into patches and discoloring to avellaneous or wood brown, the disc at times sordid alutaceous, at maturity entire surface off-color toward cinnamon buff or disc brownish (near cinnamon brown), margin fibrillose-appendiculate to cottony-appendiculate at first but finally nearly naked; flesh firm and hard (for this genus), hygrophanous, watery avellaneous fading to whitish, odor faintly fragrant but soon fading, taste not distinctive; lamellae depressed-adnate, seceding, close, narrow to moderately broad, white to very pale avellaneous becoming drab gray to hair brown, edges white-floccose; stipe 3–5 cm. long, 10–12 mm. thick, firm and hard, stuffed but becoming hollow, white within but pinkish to orange-pink in the base, surface usually with discolored fibrils over lower half, in others white fibrillose, upper half white and longitudinally striate.

Spores 7.5–9 × 4–4.5 μ, smooth, dark cocoa-color revived in KOH, narrowly oval to suboblong in face view, in side view the ventral line slightly concave and dorsal line slightly convex, apical pore present but inconspicuous; basidia four-spored, hyaline in KOH, 28–34 × 5.5–7 μ, narrowly clavate; paraphyses basidioid; pleurocystidia rare to scattered and mostly imbedded, of two types, the first clavate to subvesiculose-pedicellate, 25–40 × 10–14 μ, thin-walled, smooth and hyaline in KOH, the second type fusoid and 40–53 × 9–14 μ, thin-walled, hyaline in KOH, smooth, apices obtuse to subacute; this type varying to submucronate, when fresh often filled with oil droplets; cheilocystidia mostly similar to fusoid type of pleurocystidia or more ovate-pointed, a few clavate to subcylindric, 32–48 × 9–14 μ, hyaline and smooth in KOH; gill trama hyaline in KOH; pileus trama with a thick (300 μ) layer of veil hyphae bearing clamps, this layer very loosely interwoven and all but surface elements (which are discolored) hyaline in KOH, beneath this a cuticle of compactly arranged cells appearing vesiculose to pseudoparenchymatic in cross section (these hyaline in KOH), tramal tissue beneath cuticle hyaline in KOH.

Habit, habitat and distribution: Cespitose on dead alder wood (stumps, logs, etc.), July 28, lower Tahoma Creek, Smith *29505* —**type.** Additional collections from the same locality are as follows: July 23 (Sm. *29308*); August 7 (Sm. *29916*); August 20 (Sm. *30360*); August 22 (Sm. *30421*); August 25 (Sm. *30591*); August 27 (Sm. *30687*).

Discussion: The distinguishing features of this fungus are the innately white fibrillose caps of button stages, the fibrils or scales

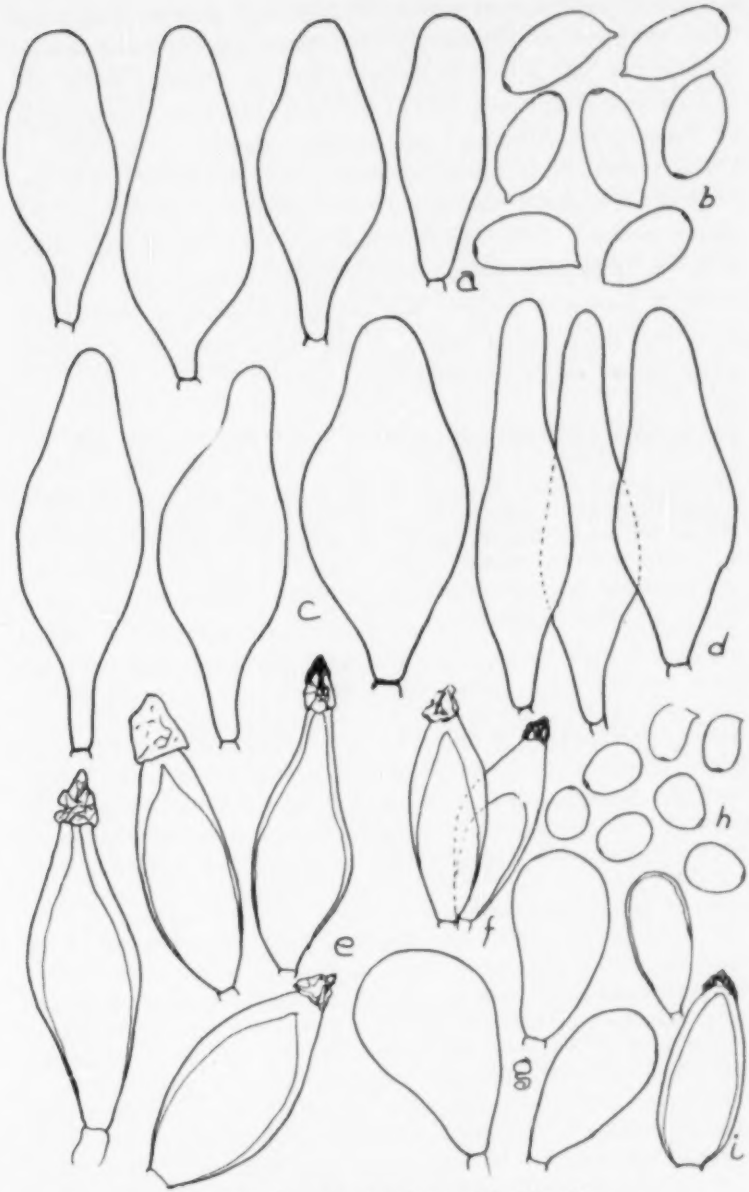

FIG. 18. Spores and cystidia of *Psathyrella*.

discoloring to alutaceous at maturity or in age, the spores (7.5–9 μ long), the two types of pleurocystidia, the pinkish to pinkish orange discoloration in the base of the stipe, and the clustered habit. It appears to be most closely related to *P. maculata*, a species described by Parker from Atkinson's Mt. Rainier collections. It differs in having larger spores and different pleurocystidia as well as in the color of the scales on the cap. In *P. maculata* the young caps are nearly fuscous. Both have been found growing on the same alder stub. It is unquestionably closely related to *Hypholoma scobinaceum* of European authors, but the gills are never tinged flesh color, and the pileus is never fuscous, unless very old discolored specimens are taken into account.

Psathyrella candidissima Smith sp. nov. *Figs.* 16*g, h;* 18*a*

Pileus 2–4 cm. latus, obtuse conicus demum subumbonatus, albo-fibrillosus demum squamulosus, glabrescens, candidus; lamellae confertae, angustae, adnatae, secedentes, candidae demum subfuscae ("light drab"); stipes 5–10 cm. longus, 4–6 mm. crassus, aequalis, cavus, candidus, floccoso-fibrillosus, subglabrescens; sporae 8–10.5 × 4–5 μ; pleurocystidia 32–40 × 10–15 μ, ventricosa, apicibus late rotundis.

Pileus 2–4 cm. broad, obtusely conic at first, expanding to nearly plane or with a low obtuse umbo, surface at first coated with a layer of snow-white fibrils more or less radially arranged and which become aggregated into fascicles before disappearing entirely, surface glabrous in age, snow-white beneath the fibrils when young, scarcely changing color in age or only the disc becoming cream color, the margin appendiculate from remains of the veil; flesh very brittle but also soft, watery pallid, fading to white, odor and taste not distinctive, no color change when bruised; lamellae close, thin, narrow, ascending-adnate and soon seceding, snow-white, becoming "light drab," edges even; stipe 5–10 cm. long, 4–6 mm. thick at apex, equal, hollow, fibrous, snow-white throughout, surface floccose-fibrillose from veil and at first with a slight evanescent zone near apex or above middle, floccose-pruinose above the zone.

Spores 8–10.5 × 4–5 μ, smooth, subelliptic in side view, in face view elliptic to subovate and broadest near base, chocolate color revived in KOH, pore apical but indistinct; basidia four-spored, hyaline in KOH, 18–23 × 7–8.5 μ; paraphyses basidioid; pleurocystidia scattered to abundant, ventricose with short broad necks and broadly rounded apices, 32–40 × 10–15 μ, smooth, thin-walled, hyaline in KOH; cheilocystidia abundant, hyaline, fusoid-ventri-

cose, apices obtuse, 32–50 × 8–12 μ, some small inflated to clavate cells also present; gill trama hyaline in KOH, subhymenium cellular; pileus trama hyaline in KOH, cuticle of a layer of pseudoparenchymatic cells only somewhat larger than diameter of cells of the flesh.

Habit, habitat and distribution: Gregarious on alder debris, lower Tahoma Creek, about 100 yards below the old Tahoma Creek Forest Camp, August 6, Sm. *29871*—**type.**

Discussion: The outstanding features of this species are the lack of an annulus, medium sized spores and broadly rounded pleurocystidia. Although found but once the species is so outstanding that it merits description. A somewhat similar species, which occurs in the Cascades farther south, has a very fragrant odor and varies in the presence of an annulus. In some respects *P. candidissima* resembles *P. insignis* but the spore size, 6.2–7.5 × 3.2–3.8 μ as compared to 8–10 × 4–5 μ, distinguishes them at once. *P. candidissima* belongs in the subgenus *Hypholoma*. *Hypholoma cascum* as described by Fries is quite similar in many respects, but Fries did not emphasize that his plant was shining white over all, in fact he described it as "griseo-alutaceo-albicans" on pileus and the gills "e griseo nigrofuscis." He does not attribute to it either a fragrant odor or bitter taste. This is important in view of descriptions by later authors. Fries' description in Monographia (p. 426) puts even more emphasis on the color of the pileus. Consequently it does not seem justifiable to identify the Mt. Rainier fungus with his species. The difference in width of the gills as expressed in our description and in all those by Fries (narrow as contrasted to "perlatis") would appear to be an additional distinguishing character.

Psathyrella caput-Medusae (Fr.) comb. nov.* *Figs.* 18*b, c, d;* 19

Pileus 4–5 cm. broad at base, nearly 3 cm. high, obtusely campanulate, surface at first covered with small whitish superficial fibrillose scales which show a tendency to become fuscous or bister at the tips, glabrescent, surface dark snuff-brown to pale snuff-brown on disc, near sepia toward the margin, hygrophanous and fading in streaks to give marginal area a coarsely striate appearance,

* *Agaricus caput Medusae* Fries, Epicr. p. 216. 1838.

near cinnamon buff faded; flesh firm but fragile, near snuff-brown moist, fading to near avellaneous, taste mild, odor sweetish aromatic; lamellae close to crowded, broad, ascending adnate, avellaneous becoming wood brown, edges even to slightly crenulate; stipe 8–10 cm. long, 7–8 mm. thick at apex, equal, hollow but not exceptionally fragile, cavity lined with avellaneous tissue, cortex

Photo A. H. Smith

FIG. 19. *Psathyrella caput-Medusae.*

paler (whitish), surface white and densely fibrillose-squarrose scaly below the cottony-membranous, fringed annulus, scales white but tips fuscescent, pruinose-silky-striate above the superior ring.

Spores 9–11.5 × 4.5–6 μ, bright cocoa-color revived in KOH but darkening somewhat on standing, subinequilateral to subelliptic in side view, apiculus distinct, suprahilar depression slight, in face view broadest near the ovate-pointed base, evenly tapered to a rounded apex, apical pore not visible under oil; basidia clavate, four-spored, 23–28 × 9–10 μ, hyaline in KOH; pleurocystidia 36–54 × (12) 14–20 (22) μ, hyaline in KOH, broadly ventricose with a short neck ending in an obtuse apex, thin-walled, many with numerous minute oil droplets; cheilocystidia similar to pleurocystidia or narrower and more elongated, 50–65 × 9–13 μ, the neck often drawn out and flexuous, apices obtuse, some with oily content, some homogeneous, thin-walled and hyaline in KOH; gill trama parallel, hyaline in KOH or at first flushed cinnamon; pileus trama parallel, hyaline in KOH or soon fading (flushed cinnamon when first revived), cuticle a layer of pseudoparenchyma 3–5 cells deep, the cells not greatly enlarged.

Habit, habitat and distribution: Cespitose on a conifer stump near Reflection Lake, September 3, collected by Stuntz (Sm. *30907*).

Discussion: The diagnostic features of this species are the habitat on conifer wood, the sweetish-aromatic odor, the darkening veil remnants, obtuse pleurocystidia many of which have oily contents, spore size, and very obscure apical pore of the spore. We have a somewhat similar species in our western mountains with a white pileus and different habitat which will be considered in a later paper. According to our observations *P. caput-Medusae* is an exceedingly rare fungus in North America. We know it from only this one collection. Morgan (**1908**) recognized it on the basis of its being reported from the Pacific Coast but ascribed spores 16–18 × 5 μ to it. We have not examined specimens, and exclude this report on the basis of the description. The Friesian account (Epicr. p. 216) describes the essential features of our collection. Lange's description and illustration cover our material even better. It remains to be seen whether the oily content of some of the cystidia is a character of importance. At present we do not so regard it. It seems apparent that the species has been variously treated by European authors. Rea (**1922**) classifies it as to group in having

an innately fibrillose pileus but in his description says it soon becomes smooth. He gives the spore size as 8–9 × 4 μ and describes the veil remnants as fuscous, not fuscesent. Bresadola states that it has no distinct odor, but in other respects his account covers our collection quite well. Through the courtesy of Seth Lundell, Upsala, Sweden, we have examined a collection by B. Norkrans from conifer wood near Upsala. The material was collected November 19, 1945 and identified by Lundell as *Hypholoma caput-Medusae*. Our Mt. Rainier collection is similar to this collection. In both the apiculus, obscure germ pore, and shape of the spore are identical as are also the pleuro- and cheilocystidia, thick cuticle of the pileus, and lack of color in the flesh of the pileus when the latter is revived in KOH. The species finds a logical place in *Psathyrella* among the annulate species of the subgenus *Hypholoma*.

Psathyrella Naucoria Smith sp. nov. *Fig.* 18*e, f, g, h, i*

Pileus 1–3 cm. latus, late convexus demum planus, margine incurvo, demum recurvo, glaber, hygrophanus, cinnamomeo-brunneus dein pallide argillaceus; lamellae pallidae demum cinnamomeae vel cinnamomeo-purpureae, confertae, angustae, late adnatae; stipes 1–2.5 cm. longus, 2–3 mm. crassus, glaber, cartilagineus, pallide ochroleucus demum subargillaceus; sporae in cumulo "Verona brown," 5–5.8 (6) × 3.5–4 × 4–4.7 μ, compressae; pleurocystidia 28–36 × 8–14 μ, fusoide ventricosa vel ventricosa-mucronata, crassotunicata, apicibus incrustatis.

Pileus 1–3 cm. broad, broadly convex with an incurved margin, becoming plane or margin uplifted, glabrous, when moist striatulate on margin, hygrophanous, "cinnamon brown," moist, fading to "cinnamon buff" or "pinkish buff," fading on the disc first; flesh thin but relatively firm, pallid, no odor or taste; lamellae pallid young, soon tinged cinnamon and becoming dark cinnamon-brown, finally with a purplish red tinge, close to crowded, narrow, horizontal and bluntly adnate, becoming shallowly adnexed, edges even; stipe short, 1–2.5 cm. long, 2–3 mm. thick at apex, usually curved, glabrous (no veil on buttons), stuffed with a pallid to white pith, distinctly cartilaginous-pliant rather than fragile, surface pruinose above, pallid honey-color young, cinnamon buff or darker in age, base slightly mycelioid.

Spores "Verona brown" to near "warm sepia" in deposits, very pale under the microscope in H_2O mounts of fresh material, when revived in KOH immature spores pale cocoa-gray, 5–5.8 (6) × 3.5–4 × 4–4.7 μ, at least some slightly compressed, broadly subovate to subglobose in face view, subelliptic in side view, pore apical

but minute; basidia four-spored, 15–17 × 5.5–6.3 μ, hyaline in KOH; paraphyses basidioid; pleurocystidia abundant, 28–36 × 8–14 μ, fusoid-ventricose to ventricose-mucronate, thick-walled at least above and mucro usually incrusted with an exudate, hyaline in KOH; cheilocystidia similar to pleurocystidia or thin-walled and vesiculose to clavate, 26–34 × 10–16 μ, hyaline in KOH; gill trama parallel or nearly so, hyaline in KOH; pileus trama hyaline in KOH, cuticle of an irregular palisade of clavate cells from between many of which clavate to sub-cylindric pilocystidia arise, these sometimes elongated into filaments, clamp connections present at cross walls.

Habit, habitat and distribution: Gregarious to scattered on very decayed alder logs, lower Nisqually River, August 3, Sm. *29785*—**type.** Coll. Sm. *30328* was made from the same log August 18.

Discussion: This species closely resembles **Psathyrella camptopoda** (Pk.) Smith comb. nov.* in habit, habitat on hardwood logs, pale spores under microscope, thick-walled pleurocystidia and stature. It differs chiefly in its much broader (in face view) slightly compressed spores. The color of the spores in KOH clearly indicates its proper position in *Psathyrella* rather than in *Naucoria*. The purplish red tinge of the gills develops slowly so that it is not too reliable as a generic character.

Psathyrella rubicola Smith sp. nov. *Figs. 20a, b, c, d, e*

Pileus 2.5–4 cm. latus, convexus vel subconicus, demum planus, albo-fibrillosus demum fibrilloso-squamulosus, glabrescens, margine appendiculatus, hygrophanus, sordide luteo-brunneus dein pallide argillaceus; lamellae pallidae ("tilleul buff") demum pallide fuscae, confertae, latae, late adnatae, secedentes; stipes 4–5 cm. longus, 3–5 mm. crassus, cavus, fragilis, candidus, fibrillosus, glabrescens et aquose pallidus; sporae 7.5–9 × 4–4.5 μ; pleurocystidia 46–60 × 9–14 μ, acuta, in KOH pallide vinaceo-tincta.

Pileus 2.5–4 cm. broad, ovoid to convex, becoming obtusely conic to convex or finally plane, surface at first covered with white fibrils which soon are grouped into fascicles and disappear (over disc first), margin conspicuously fibrillose-appendiculate from remains of the thick, fibrillose veil, surface beneath fibrils "tawny olive" but eventually darkening to drab from the spores, margin translucent-striate in age before fading, fading to near cinnamon buff or paler; flesh watery, very fragile, no odor; lamellae "tilleul buff" becoming "wood brown" to paler fuscous, close, broad, broadly adnate, readily seceding, edges even; stipe 4–5 cm. long,

* *Agaricus camptopus* Peck, Ann. Rep. N. Y. State Mus. **53**: 845. 1900.

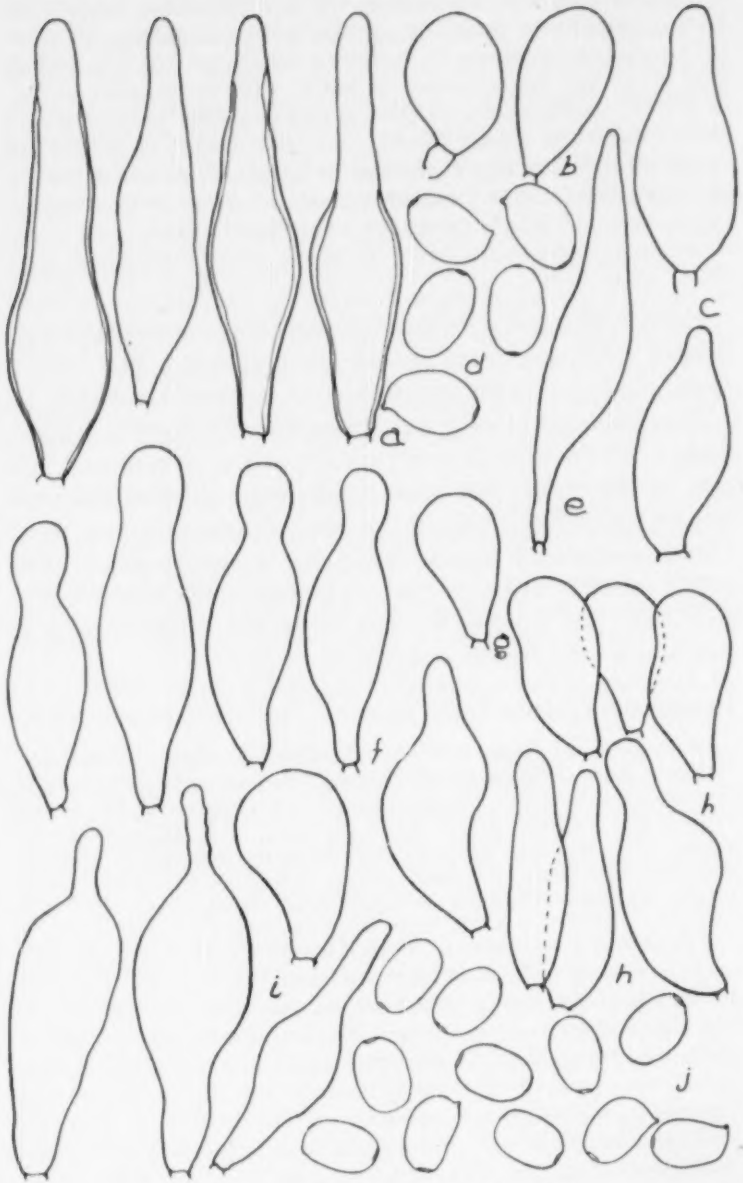

FIG. 20. Spores and cystidia of *Psathyrella*.

3–5 mm. at apex, enlarged slightly downward, hollow, very fragile, white at first because of the dense fibrillose covering, usually with a superior fibrillose zone from broken veil, also fibrillose-floccose above the zone, watery pallid to grayish beneath the fibrils.

Spores 7.5–9 × 4–4.5 μ, ellipsoid, smooth, terete, chocolate color revived in KOH, apical pore small but distinct; basidia hyaline in KOH, four-spored, 20–24 × 8–9 μ; paraphyses basidioid; pleurocystidia 46–60 × 9–14 μ, very abundant, fusoid-ventricose with acute apices, as revived in KOH with slightly thickened walls and hyaline to faintly vinaceous in KOH, the walls often flexuous, apices smooth; cheilocystidia of two types, clavate to vesiculose and up to 12–14 μ broad, these thin-walled and hyaline or faintly yellowish only at the base, second type fusoid-ventricose with acute apices, 28–40 × 9–15 μ, thin-walled, smooth and hyaline in KOH; gill trama pale cinnamon and soon fading to hyaline in KOH; pileus trama in KOH tinged cocoa-brown at first, becoming bright rusty brown just under the cuticle and paler elsewhere after standing a few minutes, cuticle a layer of vesiculose cells several cells deep.

Habit, habitat and distribution: Single or in groups of 2–3 carpophores on decaying canes and roots of Rubus species (occasionally along rotten alder logs, but here the possibility of a connection to Rubus could not be ruled out), lower Tahoma Creek about a mile above the old Tahoma Creek Camp Ground, July 19 (Sm. *29142*—**type**). Additional collections are Sm. *29148* (1 carpophore) and Imshaug *2070* (1 carpophore).

Discussion: This species is distinguished from all other species of *Psathyrella* known to me by the combination of spore size (7.5–9 × 4–4.5 μ) and acute pleurocystidia which as revived in KOH have slightly thickened walls and are frequently tinged vinaceous. There are at least two other species in North America with pleurocystidia similar to those of *P. rubicola* but both have distinctly smaller spores. They will be discussed in a future paper. Macroscopically the heavy, fibrillose, white veil and habitat are distinctive. I suspect the species of being typically vernal in its seasonal fruiting habits as it was apparently near the end of its fruiting cycle when first discovered. The species belongs in the subgenus *Hypholoma*.

Psathyrella subalpina sp. nov. *Fig. 20f, g, h, j*

Pileus 2–4 cm. latus, obtusus demum late convexus vel planus, saepe subumbonatus, canescens, glabrescens, hygrophanus, subspadiceus demum subcinnamomeus; lamellae confertae, adnatae, angustae demum latae, avellaneae demum purpureo-cinnamomeae; stipes 3–6 cm. longus, 4–6 mm. crassus, sursum attenuatus deorsum pallide fibrillosus, sursum pruinosis, brunneus, deorsum fuscescens; sporae 6–7 × 4–4.5 μ; pleurocystidia 34–46 × 8–12 μ, ventricosa, apicibus subcapitatis vel obtusis.

Pileus 2–4 cm. broad (probably larger when fully mature), buttons obtuse and expanding to convex or plane, occasionally with a broad low umbo, surface at first hoary to near center from a thin coating of veil fibrils, glabrescent over all except the incurved margin which retains a zone of avellaneous fibrils, surface moist and hygrophanous, "russet" at first but shading to cinnamon in fading; flesh thick, watery cinnamon-brown, taste none, odor imperceptible or very faint and reminding one of cinnamon; lamellae close, adnate, narrow to moderately broad, near avellaneous in buttons, changing through wood-brown to a dark purplish-cinnamon-brown, edges even; stipe (2) 3–6 cm. long, (3) 4–6 mm. diam., somewhat enlarged downward and with a mycelioid base sunken into the soil, surface pallid-fibrillose from the pale avellaneous veil, pallid and pruinose near apex, hollow, brownish within and cinnamon-brown in the base, lower half glabrescent, in old carpophores darkening from base upward.

Spores 6–7 × 4–4.5 μ, chocolate-color revived in KOH, smooth, very slightly bean-shaped in side view, in face view oblong to obscurely truncate-oblong, apical pore distinct; basidia four-spored, 20–24 × 6–7.5 μ, hymenium in thin sections hyaline, tinged cinnamon-yellowish in thick sections; paraphyses basidioid; pleurocystidia abundant, thin-walled, hyaline and smooth in KOH, 34–46 × 8–12 μ, ventricose with a narrowed neck and subcapitate apex but apex typically obtuse rather than broadly rounded; cheilocystidia of two types, the first similar to pleurocystidia, the second clavate to subsaccate and 12–18 × 7–12 μ, thin-walled, hyaline; gill trama parallel or nearly so, tinged cinnamon when revived in KOH, but soon fading, hyaline in mounts of fresh material in H_2O and KOH; pileus trama dull cinnamon revived in KOH, hyaline or nearly so in fresh material as mounted in H_2O, brownish in KOH, cuticle a layer of elongated to vesiculose cells arranged in more or less upright chains, the vesiculose cells usually the two rows nearest the surface, yellow in water mounts of fresh material, cinnamon in KOH, when revived in KOH dark cinnamon brown to russet.

Habit, habitat and distribution: Scattered on soil forming face of bank along trail, roots of conifers close by, elev. ± 4500 ft., September 10, collected by D. E. Stuntz (Sm. *31105*—**type**). Additional collections are: Sm. *30166*, August 15, and Sm. *30646*, August 26, all from the same spot as the type.

Discussion: The small spores and avellaneous veil distinguish this species, but the most interesting character is the structure of the cuticle of the pileus. In nearly all members of the subgenus

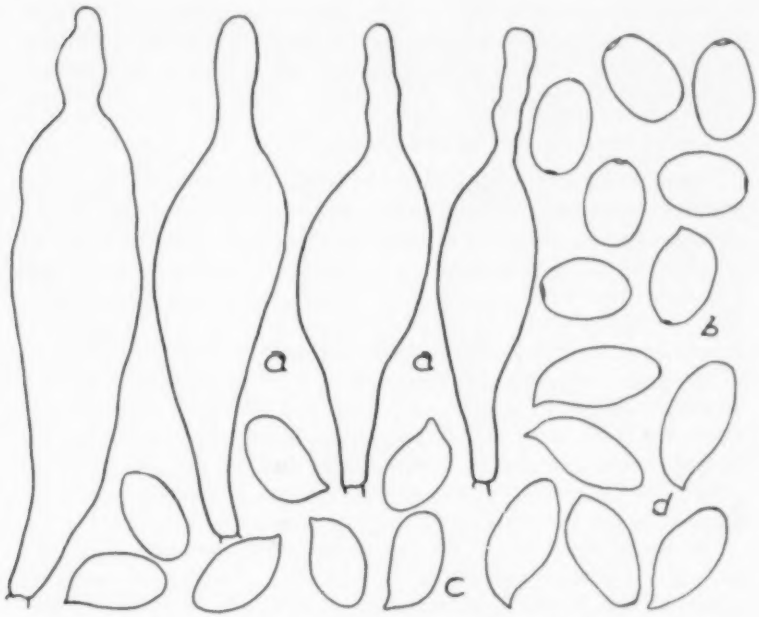

FIG. 21. Spores and cystidia of *Psathyrella* and *Clitocybe*.

the cells of this layer are hyaline either in water or revived in KOH, but in this species they are colored and when revived in KOH become very dark rusty brown much as in the cells forming the surface layer of the pileus in *Cystoderma amianthinum*. Although no globose cells become dusted over the pileus in *P. subalpina*, the manner in which the cuticle forms is quite similar in both species. If, in *P. subalpina*, the hyphae which produce the cuticular cells underwent a larger number of divisions and the re-

sulting chains of cells broke up at least in their terminal portions into individual cells, we would have the cystoderma-type of cap covering. This is of interest to students of phylogeny within the Coprinaceae because of a large number of species in *Coprinus* and a few in *Pseudocoprinus* which do have granulose pilei—the granules supposedly derived in a large part from the tissue of the universal veil.

Psathyrella subtenacipes Smith sp. nov. *Figs. 20i; 21a, b*

Pileus 1–2.5 cm. latus, obtuse conicus demum subumbonatus vel planus, udus, hygrophanus, pallide cinnamomeo-brunneus deinde pallide argillaceus, margine appendiculatus; lamellae confertae vel subdistantes, late adnatae, latae, pallidae demum subfuscae; stipes 3–4.5 cm. longus, 3–3.5 mm. crassus, deorsum subincrassatus, cavus, subtenax, albidus, fibrillosus; sporae 7.8–9.3 × 5–5.6 μ; pleurocystidia *P. delineatae* similis.

Pileus 1–2.5 cm. broad, obtusely conic, expanding to plane or slightly umbonate, surface moist and hygrophanous beneath a coating of white universal veil fibrils so thin it does not obscure the ground color, ground color pale cinnamon brown to clay-color moist, fading to cinnamon-buff on the disc and pinkish buff elsewhere, glabrescent or remaining slightly silky, margin appendiculate at first; flesh very thin and fragile, watery brownish fading to nearly pallid, odor none; lamellae close to nearly subdistant, bluntly adnate, moderately broad, pallid young, near hair brown in age; stipe 3–4.5 cm. long, 3–3.5 mm. at apex, slightly and evenly enlarged downward, hollow, cortex pallid and fibrous, *not distinctly fragile for this genus*, white but dull, more or less evenly appressed fibrillose from the remains of the veil (no zones or scales).

Spores 7.8–9.3 × 5–5.6 μ, chocolate-color revived in KOH, smooth, elliptic to subelliptic in side view, pore apical and very small, in face view elliptic to ovoid; basidia 26–30 × 7–8.4 μ, narrowly clavate, four-spored, hyaline in KOH, paraphyses basidioid; pleurocystidia 50–70 × 10–15 μ, abundant, ventricose-mucronate but mucro elongating into a flexuous almost filamentose tip with an obtuse apex, ventricose portion usually widest above the middle and tapered to the base which arises in the subhymenium or gill trama, with a large oil drop when fresh material is mounted in H_2O but homogeneous and hyaline in KOH, smooth as revived in KOH but apex with amorphous material incrusted on it as seen in water mounts of fresh material; cheilocystidia similar to pleurocystidia or smaller, some clavate to vesiculose cells present; gill trama hyaline in KOH; pileus trama hyaline in KOH, the cuticle of clavate to vesiculose cells 2–3 cells deep.

Habit, habitat and distribution: Single to gregarious on debris among willows and cottonwood, lower Nisqually River above junction with Kautz Creek, August 3, Sm. *29757*—**type.** Only the one collection preserved.

Discussion: Single specimens of this species were encountered and found not to "run down" in my key. Since most were old it was assumed that the cystidia were abnormal. When young and freshly matured specimens were finally obtained it was realized that the species was one not previously encountered. Unfortunately, after that no more material was found. The shape of the cystidia places this species beside *P. delineata*, but there is little resemblance between the two in general appearance. A rather good field character, for any one well acquainted with the fragile nature of nearly all the smaller species of *Psathyrella*, is the relatively pliant stipe. By virtue of the appendiculate cap margin the species logically belongs in the subgenus *Hypholoma*. Specimens which have lost all traces of a veil would be sought for in *Eupsathyrella*, but there are no species known to me in this subgenus which have this type of pleurocystidium.

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DESCRIPTION OF FIGURES

The drawings were made with the aid of a camera-lucida. The cystidia are reproduced at approximately 750 × and the spores 1650 ×.

FIG. 3. *a*, Spores of *Cortinarius subtortus*; *b*, spores of *Mycena rainierensis*; *c*, pleurocystidia and cheilocystidia of *Cortinarius subtortus*; *d*, cheilocystidia of *Mycena rainierensis*; *e*, spores of *Mycena fallax*; *f*, pleurocystidia of *Mycena fallax*; *g*, pleurocystidia of *Psathyrella alboalutacea*; *h*, spores of *Psathyrella alboalutacea*.

FIG. 5. *a*, Six spores of *Inocybe armoricana*; *b*, cheilocystidia of *I. armoricana*; *c*, four spores of *I. fastigiata* f. *alpestris*; *d*, cheilocystidia of *I. fastigiata* f. *alpestris*; *e*, eight spores of *I. griseo-lilacina*; *f*, three pleurocystidia of *I. griseo-lilacina*; *g*, four pleurocystidia of *I. laetior*; *h*, spores of *I. laetior*; *i*, spores of *I. leiocephala*; *j*, pleurocystidia of *I. leiocephala*.

FIG. 10. *a*, Pleurocystidia of *Inocybe oblectabilis* f. *decemgibbosa*; *b*, five spores of *I. oblectabilis* f. *decemgibbosa*; *c*, spores of *I. picrosma*; *d*, five pleurocystidia of *I. picrosma*; *e*, spores of *I. pyrotricha*; *f*, four pleurocystidia of *I. pyrotricha*; *g*, eight spores of *I. rainierensis*; *h*, pleurocystidia of *I. rainierensis*; *i*, spores of *I. suaveolens*; *j*, pleurocystidia of *I. suaveolens*.

FIG. 16. *a*, Pleurocystidium of *Psathyrella alboalutacea*; *b*, cheilocystidia of *Psathyrella alboalutacea*; *c*, spores of *Omphalina isabellina*; *d*, pilocystidia of *Omphalina isabellina*; *e* and *f*, caulocystidia of *Omphalina isabellina*; *g*, cheilocystidia of *Psathyrella candidissima*; *h*, spores of *Psathyrella candidissima*.

FIG. 18. *a*, Pleurocystidia of *Psathyrella candidissima*; *b*, spores of *Psathyrella caput-Medusae*; *c*, pleurocystidia of *Psathyrella caput-Medusae*; *d*, cheilocystidia of *Psathyrella caput-Medusae*; *e*, pleurocystidia of *Psathyrella Naucoria*; *f*, *g*, *i*, cheilocystidia of *Psathyrella Naucoria*; *h*, spores of *Psathyrella Naucoria*.

FIG. 20. *a*, Pleurocystidia of *Psathyrella rubicola*; *b*, *c*, and *e*, cheilocystidia of *Psathyrella rubicola*; *d*, spores of *Psathyrella rubicola*; *f*, pleurocystidia of *Psathyrella subalpina*; *g* and *h*, cheilocystidia of *Psathyrella subalpina*; *i*, cheilocystidia of *Psathyrella subtenacipes*; *j*, spores of *Psathyrella subalpina*.

FIG. 21. *a*, Pleurocystidia of *Psathyrella subtenacipes*; *b*, spores of *Psathyrella subtenacipes*; *c*, spores of *Clitocybe subvelosa*; *d*, spores of *Clitocybe gomphidioides*.

FURTHER INVESTIGATIONS ON THE PRESERVATION OF MOLD CULTURES [1]

DOROTHY I. FENNELL, KENNETH B. RAPER, AND MAY H. FLICKINGER

(WITH 1 FIGURE)

INTRODUCTION

Unprecedented study has centered upon the saprophytic molds during the past several years. This has resulted in large measure from the search for new antibiotic substances and from an intensive study of so-called deterioration processes. In addition, there has been a generally quickened interest in these fungi in all fields where they occur as contaminants or where they seem to offer promise of producing desirable metabolic products. As the result of this increased study, workers in many laboratories have established culture collections of varying size and diversity. In maintaining these cultures, two considerations are uppermost, namely, (1) how to prolong viability, and (2) how to preserve morphological and physiological characteristics in unaltered form.

Much attention has been given to these matters at the Northern Regional Research Laboratory, and a considerable amount of information has accumulated since our collection of cultures was established in 1940. The purpose of this paper is to review briefly some of our experiences with different methods of culture preservation, and to consider particularly some recent results obtained with cultures preserved in lyophilized form.

Four years ago Raper and Alexander (**1945**) reported the successful application of the so-called lyophil process to the preservation of a wide variety of molds. Prior to this time, bacteriologists such as Shackell (**1909**), Hammer (**1911**), Rogers (**1914**), Swift (**1921** and **1937**), Elser, Thomas and Steffan (**1935**), and Flosdorf

[1] Paper presented to the Microbiological Section, Botanical Society of America, Washington, D. C., September 10, 1948.

and Mudd (**1935** and **1938**) had developed this technique for preserving bacteria and immune sera, and had reported the successful preservation of cultures of streptococci and other pathogenic bacteria for periods up to 16 to 18 years. Wickerham and Andreasen (**1942**) had successfully used the method for preserving yeasts. Insofar as we are aware, our laboratory was the first to utilize the method successfully for the maintenance of mold cultures. Essentially, the process consists of (1) suspending conidia or other propagative cells in sterile blood serum or some other protein-rich medium, (2) dispensing the suspension into suitable tubes or vials, (3) freezing the suspension instantaneously at temperatures well below zero (−40° to −50° C.), (4) vacuum-desiccating the suspension from the frozen state, and (5) sealing under vacuum the tubes containing the desiccated preparations.

Favorable viability tests at 20 to 24 months were reported by Raper and Alexander (**1945**) for many representative species of *Aspergillus* and *Penicillium*, for 19 species of the Mucorales belonging to 14 different genera, and for a wide variety of other molds representative of the Fungi Imperfecti and the lower Ascomycetes. Consistently negative results were obtained only from two members of the Entomophthorales, namely, *Entomophthora apiculata* and a species of *Conidiobolus*. In the same paper favorable viabilities up to 38 to 41 months were reported for a more limited series of cultures including a number of industrially important strains, and others which possessed unique cultural characteristics or were reported to be unusually short-lived. Good to excellent viability was obtained in most species, and poor viabilities, wherever they occurred, could generally be attributed to a thin initial spore suspension, or were observed to be associated with forms producing large or highly organized reproductive cells. In all cases, the cultures developing from the lyophil preparations were entirely typical of the strains under observation.

FIVE- AND SEVEN-YEAR LYOPHIL TESTS

Since that time additional viability tests have been made on these same cultures after approximately 5 and 7 years. Partial results of the latter tests are shown in table I. Results of early and inter-

TABLE I

VIABILITY OF LYOPHILIZED PREPARATIONS OF SELECTED MOLD CULTURES TESTED AT INTERVALS UP TO 85 MONTHS

Name	NRRL No.	Test No. 1		Test No. 2		Test No. 4		Test No. 6	
		Age in mo.	Viability	Age in mo.	Viability	Age in mo.	Viability	Age in mo.	Viability
Aspergillus flavus Link	693	2	+++	24½	+++	38½	+++	83½	++++
A. itaconicus Kinoshita	161	3½	++	26	++	41	+	84½	++
A. niger van Tieghem	3	4	+++	26	+++	41	+++	85	++++
A. niger van Tieghem	67	3	+++	25½	+++	40½	++	84½	++
A. niger van Tieghem	328	3	++++	25	++++	39½	++++	84	++++
A. niger (tan mutant)	P-88B	4½	+++	18	++++			70½	++++
A. oryzae (Ahlburg) Cohn	692			24½	++++	38½	++++	83½	++++
A. Sydowi (B. and S.) Th. and Ch.	P-35	2½	++++	24½	++++	38½	++++	70½	++++
A. terreus Thom	265	2	++++	24½	++++	38½	++++	83½	++++
Penicillium chrysogenum Thom	811	3½	++++	26	++++	40½	++++	85	++++
P. claviforme Bainier	1002	2½	++++	24½	++++	40	+++	68½	++++*
P. islandicum Sopp	1038	2	++++	24½	++++	38½	++++	72½	++++
P. lemoni Sopp	1042	2	++++	24½	++++	38½	++++	72½	++++
P. notatum Westling	824			18	++++	34	++++	76½	++++
P. purpurogenum var. *rubri-sclerotium* Thom	1064	4	+++	26	+++	40	+++	84½	++++
P. vinaceum Gilman and Abbott	739	2½	++	24½	+	38½	+	60	+
Gliocladium vermoeseni (Biou.) Thom	1752	2	+++	24½	+++	38½	+++	74	++
Blakeslea trispora Thaxter	1718	2½	+++	24½	+++	39	+++	83½	++
Mucor Ramannianus Moller	1559	2½	++++	24½	++++	39	++++	72	++++
Phycomyces Blakesleeanus (+) Burgeff	1554	2½	+++	24½	+++	39	+++	84	++
Phyco. Blakesleeanus (−) Burgeff	1555	2½	+++	24½	+++	39	+++	83½	++
Rhizopus delemar (Boid.) Weh. and Hanz	1472	2½	+++	24½	+++	38½	+++	75	+++
Rhizopus oryzae Went. and Pr. Geerl	395	4	+++	26	++	40½	+++	85	+++

++++ = Excellent viability
+++ = Good viability
++ = Fair viability
+ = Poor viability

* NRRL 1001 substituted for 1002.

mediate viability tests are reintroduced to provide a comparison of aged, intermediate, and newly processed cultures. Results of the five-year tests have been omitted since these were comparable in all cases with the seven-year viabilities; in fact, no marked decrease in viability was observed in any species during its entire period of lyophil preservation. Naturally, a certain degree of variability occurs. For example, two strains of *Blakeslea trispora* and two of

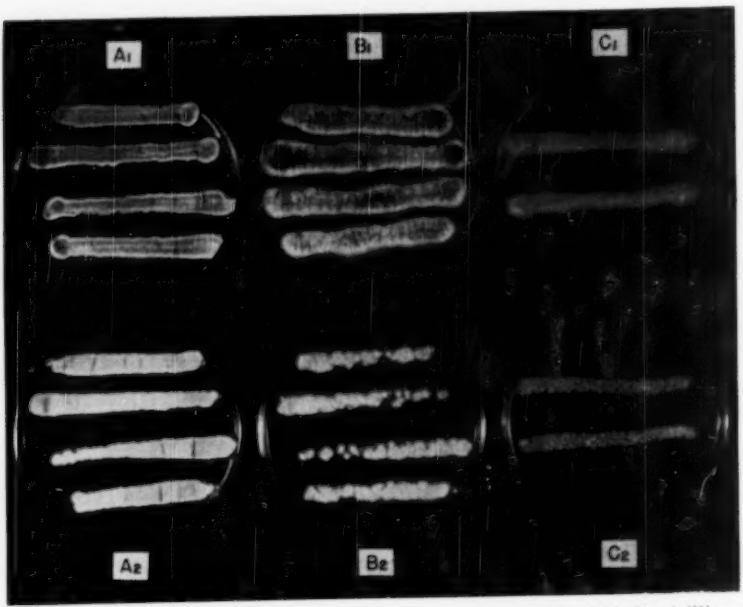

FIG. 1. Comparative growth of three representative molds, (A) *Penicillium notatum* NRRL 824 (Fleming strain), (B) *Aspergillus niger* NRRL 328, and (C) *Mucor Ramannianus* NRRL 1559, inoculated with spores from (1) current agar stocks and (2) five-year lyophilized preparations. Substrate: Czapek's solution agar. Incubaton: two days at 25° C.

Phycomyces Blakesleeanus showed only fair viability in the seven-year test after having been rated as good in all previous tests. On the other hand, cultures of *Aspergillus flavus* (NRRL 693) and *A. niger* (NRRL 3) were rated as excellent after having been scored as good in previous tests. A fluctuation in apparent viability of plus or minus 1 in our scoring system, which is admittedly arbitrary, should not be regarded too seriously. It may indicate either

a difference in evaluation of growth by different observers, or it may reflect differences in individual lyophil preparations, e.g., variation in the spore density of different fractions of the original suspension or differences in the extent of vacuum desiccation.

It is evident from the data of table I that the cultures under observation have in most cases retained their viability almost unchanged during a storage period of seven years. The viability of cultures of *Blakeslea* and *Phycomyces* may be on the decline; but simply to have maintained viable cultures of the former without transfer for seven years represents a considerable achievement. The recorded poor viability of *Penicillium vinaceum* and *Aspergillus itaconicus* is unquestionably due to the low spore content of the suspensions processed. For an *Aspergillus*, the latter species is unusually short-lived when maintained in agar slants in the conventional manner.

Whereas relatively few cultures have been preserved in a desiccated state for seven years, the entire mold collection as it then existed (numbering about 1850 different strains) was so preserved in 1942, and these preparations are now six or more years old. Tubes have been opened, as occasions demanded, and with very few exceptions viabilities have been regarded as entirely satisfactory (FIG. 1). Raper and Alexander in 1945 presented results of tests made at 1½ to 2 years on 140 representative cultures from the 1942 preservations, including 41 Aspergilli, 40 Penicillia, 29 members of the Mucorales, and 31 miscellaneous forms. In 1947 a second set of tubes from the same series of 140 cultures was opened. Results obtained in this five-year test, in the main, duplicated those previously observed. In 14 strains a marked decrease in or lack of viability was recorded, while in 4 strains the rating indicated greater viability than had been previously reported. When duplicate preparations of nine of the strains showing reduced or no viability were examined, results equal to or approaching those at the two-year interval were observed in four cases, whereas in three the decreased viabilities indicated in the primary five-year tests were confirmed. In two others, cultures were found to be nonviable: in one case (*Penicillium puberulum*) the lyophil preparation was found to be suboptimal in character with very poor vacuum and containing a soft spongy pellet—while in the other (*Chaetocladium Brefeldii*)

no physical factors which might have adversely affected viability were observed. Tests were not repeated on five of the fourteen above-mentioned strains since no preparations of the original processing date were available for testing.

COMPARISON OF DIFFERENT METHODS OF CULTURE PRESERVATION

In order to evaluate various methods of culture preservation, a rather extensive program was initiated early in 1946. The Mucorales were selected as test organisms. Methods investigated included: (1) preservation in plain agar slants, (2) preservation in fertile garden soil, (3) preservation under a mineral oil seal, and (4) preservation in lyophilized form.

The first method represented the conventional procedure practiced in most laboratories, with cultures being grown on a suitable agar medium—in this case malt extract agar. The second method represented an adaptation of a technique long employed for the preservation of bacteria, particularly the anaerobic spore-forming clostridia. As employed by us, the method followed the procedure recommended for the maintenance of molds by Greene and Fred in **1934**. With limited modifications, preservation in soil is widely employed in industrial laboratories for the maintenance of penicillin-producing molds and other economically important types. The technique of keeping cultures under mineral oil was developed for the preservation of bacteria by Ungermann (**1918**), Michael (**1921**), Morton and Pulaski (**1938**), and others, and was, apparently, first used successfully for the maintenance of fungus cultures by Sherf in **1943**. More recently it has been employed by Wernham (**1946**) and Wernham and Miller (**1948**). Conservation of fungus cultures under oil has been warmly recommended by Buell and Weston (**1947**).

In preparation for the comparative tests, four-year-old lyophil tubes of 245 cultures belonging to the Mucorales were opened and recultivated on malt extract agar. The different types of cultures used for the subsequent comparison of methods were inoculated or prepared from spores obtained in this recultivation. For each of the 245 strains, the following number and types of cultures were prepared simultaneously: two plain malt extract agar slants, a sim-

ilar slant flooded with a layer of sterile mineral oil after satisfactory growth and sporulation had occurred, a soil culture, and six lyophil preparations. All cultures were subsequently stored at 4° C.

When the cultures were somewhat more than two years old, streak plates were made on malt extract agar from all preparations, care being taken insofar as possible to use approximately equal amounts of inocula from the different types of cultures. No growth resulted from 72 of the dried-out agar slant cultures. Forty-seven of the oil-covered slants and sixteen of the soil cultures were likewise negative. In every case, however, lyophilized cultures yielded growth characteristic of the strains under observation, and in 235 strains (96 per cent), the viability was rated as either excellent or good. In four cases viability was quite poor. Good or excellent

TABLE II

VIABILITY OF MEMBERS OF THE MUCORALES CONSERVED FOR 2 TO 2½ YEARS BY DIFFERENT METHODS OF CULTURE PRESERVATION

Method \ Viability	Excellent	Good	Fair	Poor	Negative
Lyophilized preparations	205	30	6	4	0
Soil cultures	32	102	65	30	16
Oil-sealed agar slants	26	95	49	28	47
Malt agar slants	13	49	66	45	72

viability was also recorded for 135 of the soil preparations, 123 of the oil-flooded slants, and 60 of the plain agar slants. These data are summarized in table II.

Whereas no strict correlation was observable, a general relationship was found to exist between viability and species identity. Species of *Blakeslea, Chaetocladium, Circinella,* and *Cunninghamella* showed the greatest percentage of non-viability, particularly in the plain agar and in the oil-covered slant cultures. For example, of 17 strains of *Circinella* tested, only 5 remained viable under oil and an equal number, but not identical strains, on plain agar slants. Of six strains of *Cunninghamella echinata* tested, all failed to grow from the oil-covered slants, and only one of the plain agar slants yielded a viable culture. Two strains of *Chaetocladium Brefeldii* likewise failed to grow from oil-covered cultures but grew poorly from plain

agar slants, whereas the single strain of *Blakeslea trispora* included in the test proved to be non-viable in both types of cultures. In all of the above cases, good to excellent viability was obtained from lyophilized cultures of similar age.

Based upon these tests, which are summarized in table II, it would appear that the Mucorales can be maintained most satisfactorily in lyophilized form, with soil cultures, oil-covered agar slants, and plain agar slants representing progressively less favorable methods.

APPLICABILITY OF THE LYOPHIL PROCESS TO THE PRESERVATION OF OTHER FUNGI

Since the fungus cultures contained in the NRRL Collection represent, for the most part, saprophytic molds important to agriculture and the fermentation industries, our data on the applicability of the lyophil process to the preservation of fungi were largely limited to molds of this type.

Information regarding the preservation of other groups of fungi was deemed highly desirable. Additional species chosen for exploratory investigations included a limited number of aquatic Phycomycetes, representative wood-destroying Hymenomycetes, and several fungi pathogenic to man and animals. The Phycomycetes were provided by Charles Drechsler, Bureau of Plant Industry, Beltsville, Maryland; the Hymenomycetes by Ross W. Davidson, also Bureau of Plant Industry; and the pathogenic forms by C. W. Emmons, National Institute of Health, Bethesda, Maryland.

The strains belonging to each general group were cultivated on suitable media and lyophilized by our usual methods (see page 2; also Raper and Alexander, **1945**). Desiccated preparations were tested for viability immediately after processing, with the following results:

Aquatic Phycomycetes: Nine strains of *Pythium*, representing as many species, and one culture of *Plectospira myrianda* were tested with negative results. Earlier, in cooperation with Dr. John R. Raper, University of Chicago, attempts had been made to lyophilize four strains of *Achlya*, including two strains each of *A. bisexualis* and *A. ambisexualis*, in suspensions made both with blood serum

and with a thin starch paste. Negative results were obtained in these trials also and are confirmatory of those reported in Buell and Weston (**1947**) for members of this group.

Hymenomycetes: The cultures of wood-destroying fungi included three species of *Polyporus*, two each of *Poria* and *Fomes*, and one each of *Stereum* and *Lenzites*. In our cultures, all but two of these produced conidia, oidia, or chlamydospores—all representing types of spores thought to provide suitable material for lyophil desiccation. The two exceptions were *Stereum rameale* and *Fomes pinicola*. Lyophil preparations of all strains showed excellent viability except the two just mentioned and one additional strain, *Fomes annosus*. Of these, the two strains of *Fomes* showed limited viability. In three successive trials the strain of *Stereum rameale* failed to survive the lyophil process. This was considered to result probably from an observed absence of suitable propagative cells.

Pathogenic Fungi: Among the pathogenic fungi received from Dr. Emmons were seven strains of *Trichophyton*, representing four species, and one strain each of *Candida albicans*, *Sporotrichum schenckii*, *Cryptococcus neoformans*, *Blastomyces dermatitidis*, and *Histoplasma capsulatum*. Excellent growth was obtained from lyophil preparations of all but the last three strains. Of these, *Cryptococcus neoformans* showed good viability and the remaining two only fair. The apparent reduced viability of the latter may have resulted in large measure from a dearth of spores in the processed suspensions, or from the possible use of a suboptimal medium during recultivation.

The reader may justifiably question the significance of viability tests made immediately following lyophil preservation. However, we have consistently observed during an eight-year period that, once cultures have been lyophilized successfully, they retain their viability with little or no apparent decline. When difficulties are experienced, they are encountered during the freezing and drying process.

DISCUSSION

The preservation of cultures in soil, as oil-covered agar slants, or in lyophilized form necessitates the expenditure of greater initial time and effort than maintenance of the same cultures on plain

agar slants. Justification for such methods must rest, therefore, either upon prolonged viability or upon increased morphological or physiological stability. While our experience does not warrant categorical statements, it is becoming increasingly evident that among the molds producing abundant aerial spores, or conidia, viabilities are as a rule greatly prolonged by lyophil conservation. Furthermore, the new growth resulting from such cultures must develop from the actual spores or cells originally processed; hence the danger of variation is negligible except for the possibility that such may arise as a result of the freezing and drying process. It is significant that in eight years we have not observed a single instance where colonies developing from lyophilized spores differed in any detectable degree from those resulting from normal undesiccated spores of the same strain. Similarly, we have encountered no case where biosynthetic capacity has diminished as a result of lyophilization. On the contrary, in the production of itaconic acid by *Aspergillus terreus*, cultures preserved in desiccated form have maintained their capacity to produce this acid, whereas cultures maintained on agar slants by periodic transfer have shown a marked decrease in biosynthetic capacity (L. B. Lockwood, personal communication). The same may be said of the penicillin-producing species, *Penicillium notatum* and *P. chrysogenum*. However, too sweeping claims should not be made, and attention is called to the recent paper by Atkin *et al.* (**1949**), wherein lyophilization is reported to reduce the capacity of brewers' yeast to synthesize essential vitamins.

Most of the mold cultures which we maintain are well adapted for lyophil preservation since they produce abundant aerial spores or conidia. A limited number, however, cannot be satisfactorily maintained in this manner. These include forms which produce few or no conidia or other firm-walled propagative cells, and certain species which produce spores that are unusually large or highly organized. Recent experiences with certain aquatic Phycomycetes emphasize the unsuitability for lyophilization of zoospores and other reproductive structures in these forms. Repeated tests have demonstrated that vegetative mycelia, fragmented or otherwise, cannot be successfully conserved, and our experience with a limited number of wood-destroying species has confirmed this observation.

Species producing oidia or other propagative cells were successfully processed; whereas in the absence of such cells, conservation by the freeze-drying technique was not realized. The importance of cell-type to successful application of the lyophil process is beautifully illustrated by slime molds belonging to the genera *Dictyostelium* and *Polysphondylium*. Attempts to conserve the amoeboid vegetative cells have been wholly unsuccessful, whereas the capsule-like mature spores of all species tested are admirably suited for this type of preservation. Preparations up to eight years old (the maximum of our tests), if added to or preserved with a suitable bacterial host (Raper, **1937**), give rise to luxuriant and wholly typical cultures when recultivated upon suitable substrata. Such cultures develop almost as rapidly as others seeded with fresh spores.

Buell and Weston (**1947**) have correctly drawn attention to the limitations of the lyophil process as a means of conserving collections of diverse fungi and have emphasized the utility of other methods, notably conservation under oil. While we feel that they tend to over-emphasize the difficulties to be encountered in applying the lyophil process, we are in essential agreement with them and fully recognize the vital role of other methods of conservation. As a matter of fact, and depending upon the nature and importance of the strains to be preserved, any one or all of the four methods of conservation discussed above may be used advantageously. Each has its strength and its weakness, which we believe can be summarized as follows:

The agar slant method: Transfers, in whatever numbers are required, can be made quickly, and one can observe the fungus throughout its growth and development. Such cultures are not long-lived, however, and there is a constant danger of cultural or physiological variation with each period of regrowth, particularly if a degree of selection enters into the choice of the inocula used to establish succeeding transfers.

The oil-seal method: The viability of cultures is generally increased, and the method is especially applicable to the conservation of mycelial or non-sporulating forms. Mites are effectively controlled. It is receiving enthusiastic acceptance by an increasing number of investigators. However, preparation requires additional

labor and the resulting sealed cultures must be stored in an upright position at all times.

Soil preservation: Culture viability is usually substantially increased, and, if proper care is observed in handling, satisfactory and uniform subcultures can be obtained from the same preparation over long periods of time. As with the oil-seal method, preparation requires additional work, and in this case it is impossible to observe growth or to detect contamination without recultivation.

Lyophil preservation: Culture viability is greatly prolonged; new cultures arise from the spores originally processed, and the preparation of multiple tubes provides a source of uniform inoculum over an indefinite period; the possibility of contamination by mites or microörganisms is eliminated by sealing preparations in glass; strain variation is minimized through infrequent recultivation; the diminutive proportions of the finished preparations are conserving of valuable storage space. The method's most serious drawback is its inapplicability to the conservation of many types of fungus cultures which do not produce small, firm-walled spores or conidia. A less serious objection is the necessity of opening a new preparation each time a new culture of "lyophil" origin is required. Finally, preservation in lyophil form entails a greater amount of effort than any of the preceding methods.

SUMMARY

If a culture lends itself to lyophil preservation, we believe this to constitute the most reliable means of maintaining it in unaltered form over long periods of time. In the long run it is also the least time-consuming. If the culture cannot be so preserved, one must rely upon alternative methods. Preservation in soil offers certain favorable advantages, but this method—like lyophil preservation—is not applicable to the maintenance of mycelial forms. If the culture is strictly mycelial, or if very few spores are produced, preservation under oil generally affords the best means of conservation.

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MORPHOLOGY OF DISCISEDA CERVINA

SULTAN AHMAD

(WITH 3 FIGURES)

The genus *Disciseda* was proposed by Czerniaiev (**1845**) for a plant in which the exoperidium had persisted as a disc at the base. As he neither described the relevant microscopical characters nor gave any diagrams, the genus remained obscure and unrecognized for a very long time. Morgan (**1892**) described the genus *Catastoma*, characterized by the basal mouth, the capillitium, which consisted of short separate threads, and the apical collar-like remains of the exoperidium. Hollós (**1903**) dug out the generic name *Disciseda* and claimed that it was co-generic with *Catastoma* of Morgan. His labors, however, were not rewarded for a long time, as several leading mycologists preferred to use the very appropriate name *Catastoma* (basal mouth). Now, however, mycologists have begun to recognize *Disciseda* as the valid generic name on grounds of priority, even though Lloyd (**1904**) protested strongly against this tendency.

The genus is considered by several workers as curious and unique among the Gasteromycetes in having a basal mouth. Lloyd (**1918**), however, writes of *Catastoma ater* that "it is not a true *Catastoma*, . . . in the 'type idea' with 'mouth down,' but there are several species of *Catastoma* that will not stand that test." Recently Cunningham (**1946**) also states that that is not a universal feature and that in *Disciseda cervina*, *D. australis*, *D. anomala*, *D. verrucosa*, and probably others, the stoma develops apically in the usual manner. Lohwag (**1930**) finds it difficult to explain the basal mouth morphologically. The explanation offered by him is that the endoperidium at the base is torn from the "Myzelium" whereby a hole or at least a weak spot originates which later wears into an aperture.

As most of the species grow wholly or partially buried under the surface of the ground, nothing is known about the early de-

velopmental history of the fungus. The description of the various species is based only on the gross morphological features of the mature fructification. The different statements concerning the development of the mouth at the apex in several of the species are merely based on guess-work. The only reference to even the structure of the exoperidium and gleba of any species is a brief statement by Coker and Couch (1928) in connection with the description of *D. candida*.

The only representative of the genus in the West Panjab is *D. cervina* (Berk.) Hollós, which is very common in the open sandy wastes. As this species was among those in which Cunningham has reported an apical mouth, a close observation of the fungus was undertaken and an effort was made to procure young sporophores at all stages of development. Fortunately several young plants were collected in the summer of 1948, growing in a locality from which hundreds of mature specimens had been obtained in the past.

Several of these plants were fixed immediately in formalin-acetic-alcohol, the only fixative available at the time. The sections were cut 5–10 μ thick and stained with iron hematoxylin and phloxine.

OBSERVATIONS

The young sporophores are always obovate or turbinate with the apical part more or less exposed. They are attached by a well developed mycelial cord at the base. The only species in the vicinity with the sporophores of which these could be confused are *Lycoperdon pusillum* and *Lanopila wahlbergii*. The former is always epigeal and the latter, though hypogeal, has a slightly different color and avoids loose sandy soil. With a little experience in the field one can easily recognize the young sporophores of *D. cervina* by their form, color and peculiar behavior during later stages of growth.

The first thing in the young sporophores which attracts attention is the floccose mycelium arising from the exposed top portion. These sporophores when allowed to develop *in situ* disappeared from sight during the night. It was very difficult to guess what had happened to them. It was first thought that they had proba-

bly been eaten up by some rodent, but there were no fragments lying nearby to confirm this conjecture. After experiencing the same difficulty for two or three days successively a few plants were covered with small earthen cups but the result was still the same. Very much puzzled at this, the soil under the cups was carefully dug out and then it was discovered that the plants had buried themselves under the soil in all cases. A more careful observation revealed that the hyphae of the floccose mycelium at the top ramify and enclose numerous grains of sand and disappear from sight. Among two or three dozen plants unearthed at different intervals of time, it was found that the formation of the hyphal covering enclosing sand particles progresses from the apex downwards till the whole plant is covered with a sandy case. The sandy case is more or less uniform in thickness and so the plants retain their original form.

The sandy case was never seen to split in a circumscissile manner described and figured by Morgan. On the other hand, it always crumbles from the base upwards after the plants have been unearthed by wind. When most of the loose basal part is lost the heavy top assumes a basal position. In the plants unearthed after about ten days the basal mycelial cord was still intact. In some, however, it appeared to have gelatinized at the base, as it was easily pulled out, leaving a pore—the mouth or stoma—at the base.

Mycelial cord. The mycelial cord consists of a central medulla, a middle sub-cortical region, and an outer cortex. The medulla consists of thick, wide-lumened hyphae intermixed with numerous thin-walled hyphae. The sub-cortical zone is formed of very compact and closely interwoven hyphae. The outermost hyphae of the cortex branch and enclose numerous particles of sand.

Development. The young sporophores develop at the apex of a mycelial cord. Sometimes two growing close together become fused with one another. At a very early stage of development each consists of loosely interwoven hyphae continuous with those of the parent strand. The thick, wide-lumened hyphae of the medulla and the hyphae of the cortex do not take part in the formation of the young sporophore. In the center of the homogeneous tissue are later differentiated small cavities which gradually pro-

gress towards the periphery. The development corresponds in all essential respects to the *lacunar type* as described by various investigators for other members of the family Lycoperdaceae.

Exoperidium. The hyphae forming the primordial tissue radiate towards the periphery; they are parallel and wide-lumened. These hyphae become closely septate and the cells become swollen and compacted to form pseudoparenchymatous tissue. In a fully formed plant this layer is 400–500 μ thick and consists of several

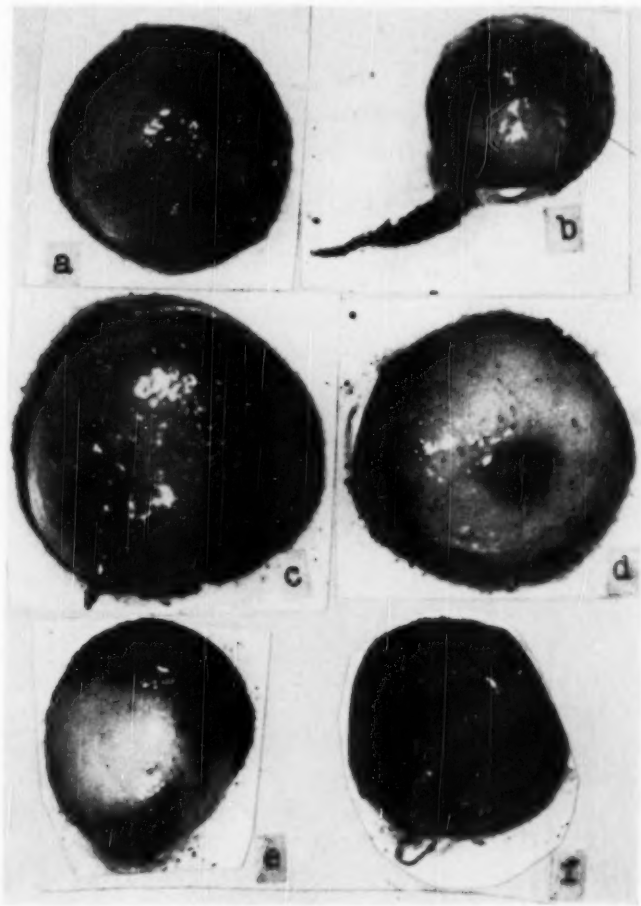

FIG. 1. *Disciseda cervina.*

layers of thin-walled cells. This layer though continuous around the upper part of the fruitbody is poorly developed at the top. At the base, however, it is interrupted by the mycelial cord.

After the pseudoparenchymatous layer has been organized, a second or mycelial layer begins to form. This is, however, completed after the differentiation of the endoperidium has taken place. The mycelial layer is composed of long, intertwined, partially gelatinized hyphae which arise from the exposed top portion of the sporophore. The hyphae ramify in the surrounding soil and enclose numerous sand particles to form a small cap at the apex (FIG. 1, *a–e*). In the course of time the mycelial layer progresses downwards, ultimately forming a complete sandy case around the sporophore (FIG. 2, *a*). It is only incomplete at the base from where the mycelial cord comes out (FIG. 1, *f*).

The pseudoparenchymatous layer is very poorly developed at the top and several hyphae of the endoperidium are also seen passing through it into the mycelial layer. On the sides below the apical disc there is absolutely no connection between the mycelial layer and the hyphae of the endoperidium. This intermixing of the hyphae of the mycelial layer and the endoperidium at the top accounts for the persistence of the apical collar or disc.

The cells of the pesudoparenchymatous layer collapse at maturity and form a thin membrane on the inner surface of the sandy case. The disorganization of this layer at maturity loosens the sandy case all round excepting at the apex, as explained above. When the plants are exposed by wind they lie scattered on the surface of the ground with the sandy case still intact. The loose basal part gradually crumbles by the attrition of sand particles until only the apical disc is left (FIG 2, *b–i*).

The top-heavy plant now easily gets overturned and the collar or the disc appears to be basal. The appearance of the "*exoperidium*" as collar or disc depends on the size of the case crumbled at the time of collection. In very old, weathered plants there is only a small disc left at the base, indicating the region where the union was strongest (FIG. 2, *j–w*).

Gleba. The hymenial cavities originate and increase in number exactly as in other members of the family. The ground tissue grows by the expansion of its elements and the formation of new

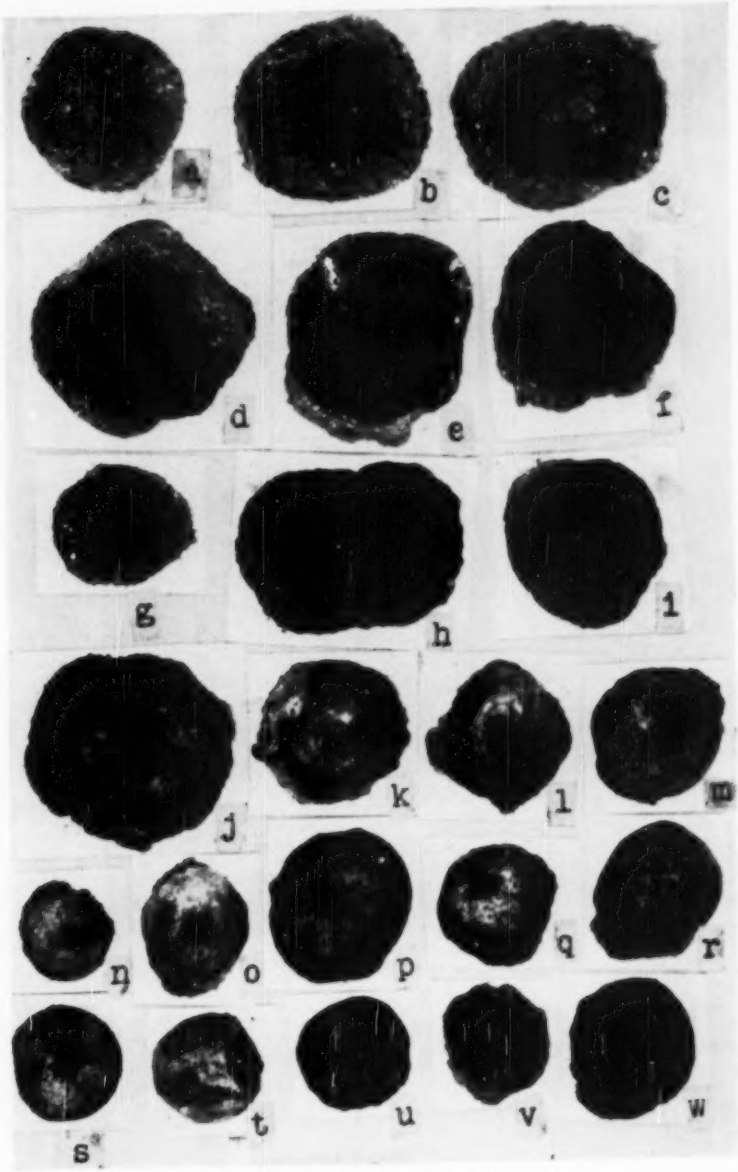

FIG. 2. *Disciseda cervina.*

tramal plates. Most often stout hyphae pass from one side of the hymenium to the other across the hymenial cavity. As pointed out by Swartz (**1933**) these hyphae clearly suggest that the formation of cavities results entirely from disintegration of the primordial hyphae.

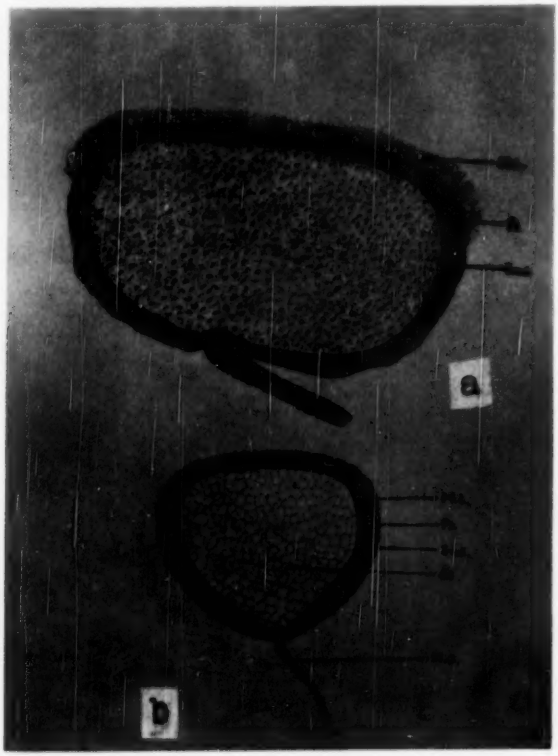

FIG. 3. *Disciseda cervina.*

The basidia, arranged in compact hymenia, are short and pyriform, bearing four spores apically on long sterigmata. The thickening on the spores develops after they have been detached from the sterigmata. After the spores are formed, a rapid disintegration of all the remaining glebal hyphae and basidia takes place, leaving nothing excepting spores and capillitium. The capillitial threads arise from the tramal hyphae and not in the manner de-

scribed by Cunningham (**1926**) for *Lycoperdon depressum*. At maturity the capillitium breaks up into short segments.

Endoperidium. When the development of the mycelial layer has not yet progressed even half way down, the endoperidium becomes differentiated (FIG. 3, *a*). It is formed immediately below the pseudoparenchymatous layer of the exoperidium. In earlier stages this region appears as an undifferentiated outer part of the gleba bordering the hymenial cavities. Later there are differentiated in this region numerous stout interwoven hyphae which become arranged tangentially and form the fundament of the endoperidium. At this time it is about 150 μ thick and the glebal tissue has not yet disintegrated.

The endoperidium is interrupted at the base by the mycelial cord, which occupies an area of about 1.16 mm. in size. When the basal part of the mycelial cord penetrating the exoperidium and the endoperidium gelatinizes, an opening is left at the base which serves as a mouth or stoma.

DISCUSSION

The genus *Disciseda* is included in the family Lycoperdaceae. According to Cunningham (**1946**, p. 28) it is "the most primitive genus, as is shown by its external sand-case exoperidium and somewhat hypogean habit." This statement is evidently not based on any ontogenetic investigation of the young stages. There is no doubt that in the lower members of the group we come across a peridium which is always a simple undifferentiated layer of the ground tissue, but in *Disciseda* the peridium consists of two distinct layers—an exoperidium and an endoperidium. The exoperidium is further differentiated into an inner pseudoparenchymatous and an outer mycelial layer.

According to Lohwag (**1928**) the number of peridial layers is not an absolute criterion of higher organization but depends on the form of the fruitbody. Still, if the peridium must be taken as of some systematic value, it will be seen that *Disciseda* is more highly organized than all the other members of the family Lycoperdaceae. The differentiation of the peridium into several

layers takes place according to varied and important ecological functions.

It may also be true, as pointed out by Gäumann and Dodge (**1928**), that the primitive Gasteromycetes are mostly hypogeous but with a strong tendency to elevate the gleba above the earth to secure the dissemination of their spores by wind or insects. In assigning *Disciseda* a primitive position on this ground, Cunningham has overlooked the most fundamental fact that the subterranean habit has developed independently among several different groups of fungi which are the most highly organized.

In the Gasteromycetes a pulverulent gleba, well developed capillitium, and a mouth for the dissemination of spores by wind are characters closely associated with an epigeous mode of life. It is, therefore, safe to assume that *Disciseda* has evolved from some epigeous ancestors and that the subterranean habit is of ecological rather than of phylogenetic interest. It appears to me that the hypogean habit, in plants growing in hot sandy areas, has developed in response to the extremely xerophytic conditions of life. As the loose sandy soil has little power to retain water and the superficial layers of soil soon become dry even after a heavy shower, the plants have secondarily retired under soil. Under these conditions, we see that the most successful forms are those which originate the deepest of all and these are *Montagnites arenarius*, *Tulostoma volvulatum*, *Schizostoma mundkuri*, *S. laceratum*, *Podaxis pistillaris* and *Phellorina inquinans*.

If we imagine a large number of species growing on the edge of a drifting sand dune and during the course of ages regularly buried under sand, it would become clearer how they acquire their present-day mode of life. At first the layers of sand were probably not thick enough or were washed away and the dispersal of spores was effected in a normal fashion. When the layers of sand became sufficiently thick only such plants escaped elimination as could produce some mechanism to elevate their sporocarp for the dissemination of spores by wind. Some plants, like species of *Geaster*, developed a highly specialized peridium, while others developed a very long woody stipe for this function. The former set of species was confined to the superficial layers of soil while

the latter was adapted to very deep layers. The woody stipe is an absolute necessity for the breaking of the thick layers of soil above the sporophore. It is interesting to find that under similar environmental conditions even a *Coprinus*-like *Montagnites* developed a long woody stipe similar to other Gasteromycetes living in the same habitat.

The same thing has been observed by the late Dr. W. H. Long (**1942**) in his study of the different species of *Geaster*. He writes as follows: "All species of *Geaster* in my territory are hypogeous when young and remain so until expansion. I have never seen a truly epigeous *Geaster* in the dry southwestern regions. The reason for this is evident, in that our climate below 8000 ft. elevation is too dry for species of *Geaster* to grow on the top of the ground as some species do in the humid regions of the north and east. This need of moisture also explains their presence under trees and shrubs where the moisture is retained until these fungi mature."

The hypogeous species originating in the superficial layers of sand stand at a distinct disadvantage as compared to those mentioned above but *Disciseda cervina* has specialized in a particular direction. The plant has developed an efficient mechanism for "burrowing" into the loose sand and at the same time forming a case of sand which serves it as a moist chamber. This minimizes the chances of sudden drought overtaking the developing sporophores. The mechanism is so efficient that of the two or three hundred individuals gathered at different times there was not even one in which the gleba had not reached proper maturity.

Another specialization consists in making use of the sandy case in the dissemination of the spores. The basal mouth which developed as a necessity is definitely a disadvantage but the heavier apical disc shifts it to the top so that it can play its normal role in the life of the plant. This is the most curious phenomenon observed in this species, and is unparalleled in the whole group of fungi.

In *Lycoperdon pusillum*, an epigeous species growing side by side with the *Disciseda*, one finds about fifty per cent of the plants with growth arrested at some stage before the proper ripening of

the gleba. The same thing has been observed in *Lanopila wahlbergii*, which though hypogeal occurs in the superficial layers of the soil and without any specialization. It is doubtful if the latter species can be classed as among the true arenicolous species mentioned above, as it avoids the loose sandy soil and seeks the shelter of the shrubs.

It has been customary to include all the Gasteromycetes with capillitium and pulverulent gleba in the family Lycoperdaceae. Different functions have been assigned to the capillitial threads by different workers. According to de Bary (**1887**) and others they help in the dispersal of spores but according to Swartz (**1933**) they are associated more with the disposal of waste products than with the dispersal of spores. Whatever their function may be in the earlier stages, one thing is certain and it is that at maturity they prevent the collapsing of the peridium when it is pressed by some external agency. They ensure thus the dispersal of spores at intervals in puffs or clouds.

The form of the capillitium varies in the different genera of the family. In *Lycoperdon* and *Calvatia* the capillitium is well developed and consists of long, branched, septate, and intertwined threads. In *Bovista* and *Mycenastrum* the capillitium consists of short separate threads. In *Disciseda*, *Abstoma*, and *Lanopila* the capillitium is of the *Lycoperdon*-type but at maturity it breaks up into short segments. The significance of the different forms of capillitial threads in one family is obscure, especially when they are supposed to undertake the same function.

The genus most closely related to *Disciseda* is *Abstoma*. The genus *Abstoma* resembles *Disciseda* in every respect excepting the absence of a mouth. The reticulate spores do not serve as a distinguishing feature, as Zeller (**1948**) has recently described a species with verrucose spores. Until the development of the genus is studied it would be difficult to state how the mouth has become eliminated.

In the present state of our knowledge, and assuming that the hypogeal forms have gradually evolved from the epigeal forms under changed ecological conditions, the genus *Disciseda* would be regarded as a very highly specialized member of the family Lycoperdaceae.

SUMMARY

1. *Disciseda cervina* is semi-hypogeal in the earlier stages but becomes completely hypogeal by developing a mycelial layer round it.

2. The sporophore is attached by a well developed mycelial cord of complex structure.

3. The development is of the *lacunar type*.

4. The peridium consists of three layers: mycelial layer, pseudo-parenchymatous layer and the endoperidium.

5. The mycelial layer forms around the entire fruit-body a sandy case which crumbles down when the plant is exposed by wind. The sandy case persists as an apical disc and helps in inverting the plant.

6. The mouth is formed at the base by the gelatinization of the basal part of the mycelial cord penetrating the exoperidium and the endoperidium. When the top-heavy plant becomes inverted the mouth assumes an apical position.

7. The structure of the gleba and the origin of the capillitium is of the same type as described for other members of the family.

8. The hypogeous habit has been secondarily acquired under changed environmental conditions.

9. The genus *Disciseda* is a highly specialized member of the family Lycoperdaceae.

The writer is deeply indebted to Dr. Nazir Ahmad and Sher Ahmad Khan Lodhi for aid in the preparation of the manuscript and to Dr. Donald P. Rogers for reading the typescript.

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EXPLANATION OF FIGURES

FIG. 1. Young sporophores showing different stages in the progression of the mycelial layer. In *f* only a small portion round the mycelial cord remains uncovered. × 2.

FIG. 2. *a*. Sporophore wholly covered with a sandy case. The basal mycelial cord was broken by careless handling. *b–i*. Mature plants showing gradual crumbling of the sandy case from the base upwards. The basal orifice is clearly seen in some. *j–tc*. Several plants collected in February 1949 showing the marked reduction in size of the apical disc due to weathering. × 2.

FIG. 3. *a*. Semi-diagrammatic section of young fructification showing the formation of the mycelial layer at the top. The endoperidium is fully differentiated at this stage. The two peridium layers are interrupted at the base by the mycelial cord. *b*. Diagrammatic section of a mature fructification. M.L. Mycelial layer; Ps. Pseudoparenchymatous layer; End. Endoperidium; Gl_t Gleba; M.C. Mycelial cord.

PRODUCTION OF HYDROCYANIC ACID BY CULTURES OF A BASIDIOMYCETE [1]

WILLIAM J. ROBBINS, ANITA ROLNICK, AND FREDERICK KAVANAGH

(WITH 3 FIGURES)

In the course of investigations on the production of antibacterial substances by Basidiomycetes, an unidentified fungus (B841) from white cedar [2] was grown on a liquid medium in 2800 ml. Fernbach flasks in a large cabinet maintained at 25° C. It was observed that *Penicillium funiculosum* grew unsatisfactorily in this cabinet on agar slopes in test tubes (FIG. 1). Investigation demonstrated that the poor growth of the *Penicillium* was caused by a volatile toxic substance produced by the B841 fungus. The toxic substance is believed to be HCN.

Although HCN was first reported in *Marasmius oreades* by v. Losecke in 1871 (**5**) and has since been demonstrated in the sporophores of a number of Basidiomycetes (**1, 2, 3, 4, 6**), the production by fungous mycelium in culture of sufficient amounts to interfere with the growth of neighboring cultures of other organisms has not been reported.

The cabinet—7 feet high, 6 feet wide and 2 feet deep—contained 7 shelves with 48 Fernbach flasks each inoculated with fungus B841 and distributed as follows: 16 on the first shelf, 6 on the second, 3 on the third, and 23 on the fourth. Each Fernbach contained 1 liter of a corn steep medium (**7**). The dry weight of the mycelium of fungus B841 in a Fernbach flask was approximately 3.5 g. and the surface area about 255 sq. cm. The cultures of the *Penicillium* were incubated on the fourth shelf. The doors of the cabinet were not hermetically sealed and were, as a rule, opened once or twice each day of the week except Saturday and Sunday. Under these circumstances, the accumulation of sufficient HCN

[1] This investigation was supported in part by the Howard Bayne Research Fund of The New York Botanical Garden.

[2] We are indebted to Dr. Dow V. Baxter for the culture of this fungus.

or other volatile toxic material to become inhibitory was unanticipated.

The inhibitory action of fungus B841 was confirmed by covering one of the cultures in a Fernbach flask with a bell jar. Agar slants inoculated with *Penicillium funiculosum* were incubated under the bell jar and duplicate cultures were incubated outside. The

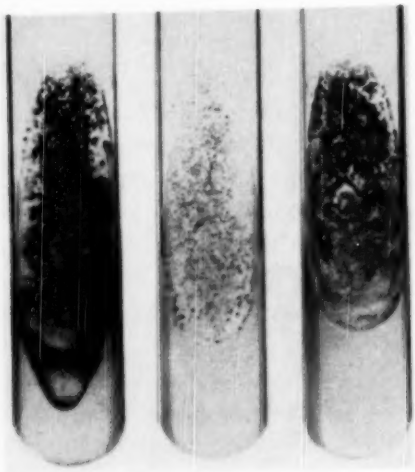

FIG. 1. *Penicillium funicolosum* grown 11 days on $NaNO_3$ agar. From left to right grown at 20° C. with no cultures of fungus B841; at 25° C. with cultures of fungus B841 in the cabinet; at 25° C. with no cultures of fungus B841.

media used were 2 per cent malt agar, thiamine-peptone agar [3] and an agar with $NaNO_3$ furnishing the nitrogen.[4] At intervals the bell jar was lifted and aerated to avoid anaerobic conditions. The cultures of fungus B841 were approximately 3 months old but the culture liquid had been decanted and replaced with fresh sterile medium at intervals of about 4 weeks. The fungus mat was heavy, white and vigorous; no fruiting bodies were present. The growth

[3] The thiamine-peptone medium contained per liter of distilled water 0.5 g. $MgSO_4 \cdot 7H_2O$, 1.5 g. KH_2PO_4, 2 g. neopeptone, 0.5 ml. mineral supplements used in this laboratory, 50 g. dextrose and 1500 mμ moles thiamine.

[4] In making the $NaNO_3$ medium the peptone was replaced with an equal weight of $NaNO_3$. The pH of the agars was as follows: malt agar pH 4.9, thiamine-peptone agar pH 5.5, $NaNO_3$ agar pH 5.3.

of *Penicillium funiculosum* was substantially reduced in the cultures under the bell jar; the effect was most marked on the $NaNO_3$ medium (FIG. 2).

A series of microörganisms was tested in the same way. It included *Penicillium notatum, Ceratostomella ulmi, Merulius lachrymans, Poria incrassata, Phycomyces blakesleeanus, Agaricus campestris, Aspergillus niger, Rhizopus nigricans, Trametes pini, Rhodotorula aurantiaca, Saccharomyces cerevisiae, Polyporus schweinitzii* and *Neurospora sitophila.*

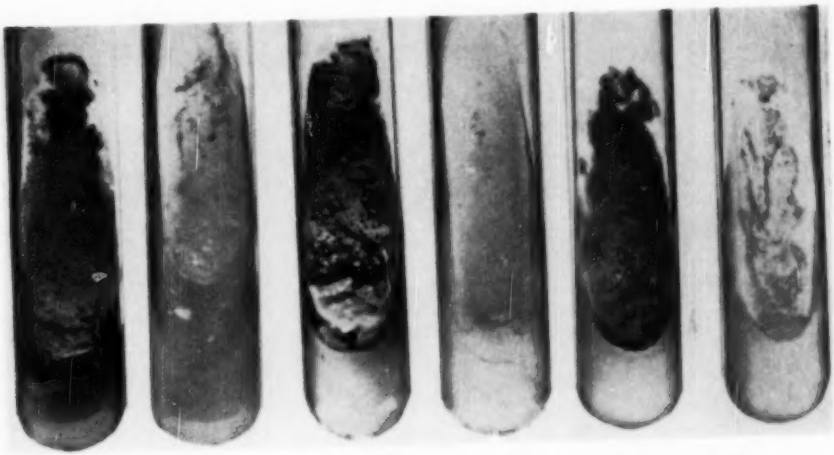

FIG. 2. *Penicillium funiculosum* grown at room temperature on various media under bell jar in presence of Fernbach flask containing fungus B841 and outside the bell jar. From left to right 11 days on malt agar outside bell jar; under bell jar; 14 days on $NaNO_3$ agar outside bell jar; under bell jar; 11 days on peptone agar outside bell jar; under bell jar.

All of the fungi tested showed some inhibition under the bell jar but the responses varied with the fungus and with the medium. Generally speaking, the inhibition was greater for growth on the $NaNO_3$ medium than for that on the malt or peptone media. Some of the organisms used grew very poorly on the $NaNO_3$ medium because of vitamin deficiencies or an unfavorable nitrogen source.

Aspergillus niger, Poria incrassata, Penicillium notatum, Polyporus schweinitzii, Neurospora sitophila and *Saccharomyces cerevisiae* were affected least. At the end of 2 weeks there was little

difference in the appearance of the cultures inside and outside the bell jar.

The mycelial growth of *Phycomyces blakesleeanus* and *Rhizopus nigricans* was substantial under the bell jars but the production of sporangiophores was materially reduced. On the malt and peptone agars *Phycomyces* formed sporangiophores in the cultures under the bell jar which failed to mature and both sporangiophores

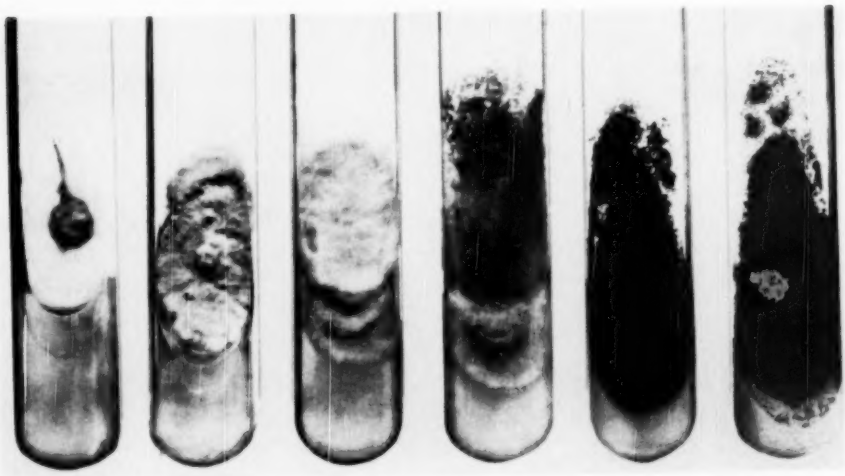

FIG. 3. Various fungi grown on thiamine-peptone agar at room temperature under bell jar in presence of Fernbach flask containing fungus B841 and outside the bell jar. From left to right *Merulius lachrymans* 26 days old under bell jar; outside bell jar; *Rhizopus nigricans* 20 days old under bell jar; outside bell jar; *Aspergillus niger* 20 days old under bell jar; outside bell jar.

and mycelium were a deep yellow. *Rhizopus* developed few sporangiophores under the bell jar.

The growth of *Ceratostomella ulmi*, *Penicillium funiculosum*, *Merulius lachrymans*, *Agaricus campestris*, *Trametes pini*, and *Rhodotorula aurantiaca* was markedly reduced. Typical responses are shown in figure 3.

Yellow pigment appeared in the agar of cultures of *Penicillium notatum* and *Merulius lachrymans* incubated outside the bell jars; pigment production was inhibited in cultures under the bell jar.

Tubes of sterile agar were incubated under a bell jar with a culture of fungus B841 for three days. The tubes were removed, inoculated with *Penicillium funiculosum*, and incubated in a cabinet containing no cultures of fungus B841. Comparable tubes which had stood outside the bell jar were inoculated and incubated at the same time. Growth was less in the tubes which had been under the bell jars for 3 days before inoculation. The effect was most marked for the $NaNO_3$ agar and least for the malt agar. This demonstrated that a volatile toxic substance produced by fungus B841 was absorbed by an agar medium incubated in the presence of a culture of the fungus under a bell jar.

The production of hydrocyanic acid by cultures of B841 was demonstrated by the Guignard test and by the Prussian blue method. Strips of filter paper impregnated with sodium picrate suspended in the air above the culture liquid of fungus B841 gave the characteristic color reaction for HCN. The contents of an open dish originally containing potassium hydroxide left in the incubator for several days gave a strong test for cyanide by the Prussian blue method. The odor of HCN was detected in the cabinet in which fungus B841 was incubated and in the bell jar which covered a culture of the fungus.

We were not successful in demonstrating the production of HCN by actively growing mycelium of fungus B841. It seems probable that the HCN originated from autolysis. Bach (1) found HCN in fruit bodies of *Pholiota aurea* (Matt.) and *Clitocybe geotropa* (Bull.) 48 hours after picking but none when they were fresh. The formation of the HCN appeared to be enzymatic and perhaps the result of the oxidative decomposition of amino acids.

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THE PHYSIOLOGY OF A BLUE STAIN MOLD WITH SPECIAL REFERENCE TO PRODUCTION OF ETHYL ACETATE [1]

MORRIS A. GORDON [2]

(WITH 9 FIGURES)

In the course of a survey of fungi causing blue stain of timber in Duke Forest, in the Spring of 1947, there was isolated from a blue-stained recently-cut stump of short-leaf pine (*Pinus echinata* Mill.) an ascomycete which produced, in culture, a strong ester odor, commonly described as banana oil odor, suggestive of the lower organic acetates. The present account is concerned with the identity of this fungus and isolation and identification of the odoriferous substance, with an attempt to discover the chemical mechanism of its formation.

Few accounts of ester production by fungi appear in the literature. Birkinshaw, Charles, and Raistrick (**4**) recovered ethyl acetate from cultures of *Penicillium digitatum* Saccardo. Birkinshaw and Findlay (**5**) identified methyl cinnamate, methyl p-methoxycinnamate, and an ester of anisic acid in cultures of *Lentinus lepideus* Fr., and Birkinshaw, Bracken, and Findlay (**3**) isolated methyl anisate from the metabolic products of *Trametes suaveolens* (Linn.) Fr. Nord and Vitucci (**12, 13**) found that *L. lepideus* produced methyl p-methoxycinnamate from various carbon sources, and attempted to elucidate the mechanism of its formation.

Several species of *Endoconidiophora* have been mentioned in connection with the formation of esters or of ester odors. Lagerberg, Lundberg, and Melin (**9**) reported "amyl acetate" produc-

[1] Based on a thesis submitted to the graduate school of Duke University in partial fulfillment of the requirements for the degree of Doctor of Philosophy.

Appreciation is hereby expressed by the author to Professor F. A. Wolf for his advice and encouragement during the course of the work and to Professor L. E. Anderson for aid in the illustrative photography.

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tion on various carbon sources by *Endoconidiophora coerulescens* Münch. According to Lagerberg (**8**) this was not established chemically, but only by odor. Morgan (**11**) found by chemical analysis that the odor produced by *E. coerulescens* Münch is caused largely by the presence of methylheptenone, although some iso-butyl acetate also was isolated. Davidson (**6**), in describing *E. moniliformis*, attributed to it an odor of banana oil and commented that "the production of a distinct odor seems to be a character common to all of the species . . . [of *Endoconidiophora*]". Davidson (**7**) states that *E. virescens* Davidson is characterized by a "musty penetrating odor" and *E. variospora* Davidson by an "odor similar to banana oil".

MATERIALS AND METHODS

Isolation of the ascomycete from the stained wood was accomplished by implantation of perithecia-bearing splinters on 2.5 per cent Difco malt extract agar plates and transfer of ascospore masses from the resulting crop of perithecia to new plates. Stock cultures bearing always both perithecia and conidia were maintained on slants of the same medium in pyrex test tubes incubated at 20° C., and transfers made at 4–5 day intervals by mass inoculation of either ascospores or perithecia. By this procedure large concentrations of perithecia were produced with a minimum of accompanying mycelium and conidia. For the inoculation of liquid media, slants were flooded with 7–8 ml. of sterile distilled water and the resulting suspension of ascospores and conidia pipetted into the media in the indicated amounts. All cultures were incubated aerobically at 20° C. (± 1°) in the dark, except as otherwise noted.

Media were sterilized at 15 lbs. for 15–20 minutes, except in the case of urea and certain other indicated heat-labile components which were filtered through a Seitz filter and added aseptically. Thiamin was autoclaved separately.

In growth measurements, where "no growth" is noted, more or less extensive germination has occurred in each case. "Inhibition" indicates lack of germination.

Dry weights were determined by filtration of liquid cultures (25 ml. of medium) through tared oven-dried Whatman No. 1 filter

paper in Gooch crucibles and drying of the fungus mats for 48 hours in a hot-air oven maintained at 100–105° C.

Ester titrations were done according to A.O.A.C. (1) procedure, on cultures grown in 250 ml. Erlenmeyer flasks containing 100 ml. of medium. In each case the mat was broken up with dissecting needles and the entire contents of the flask distilled.

Hydrogen ion concentrations were measured with a Beckman pH meter, necessary adjustments of the media being made with NaOH and H_2SO_4.

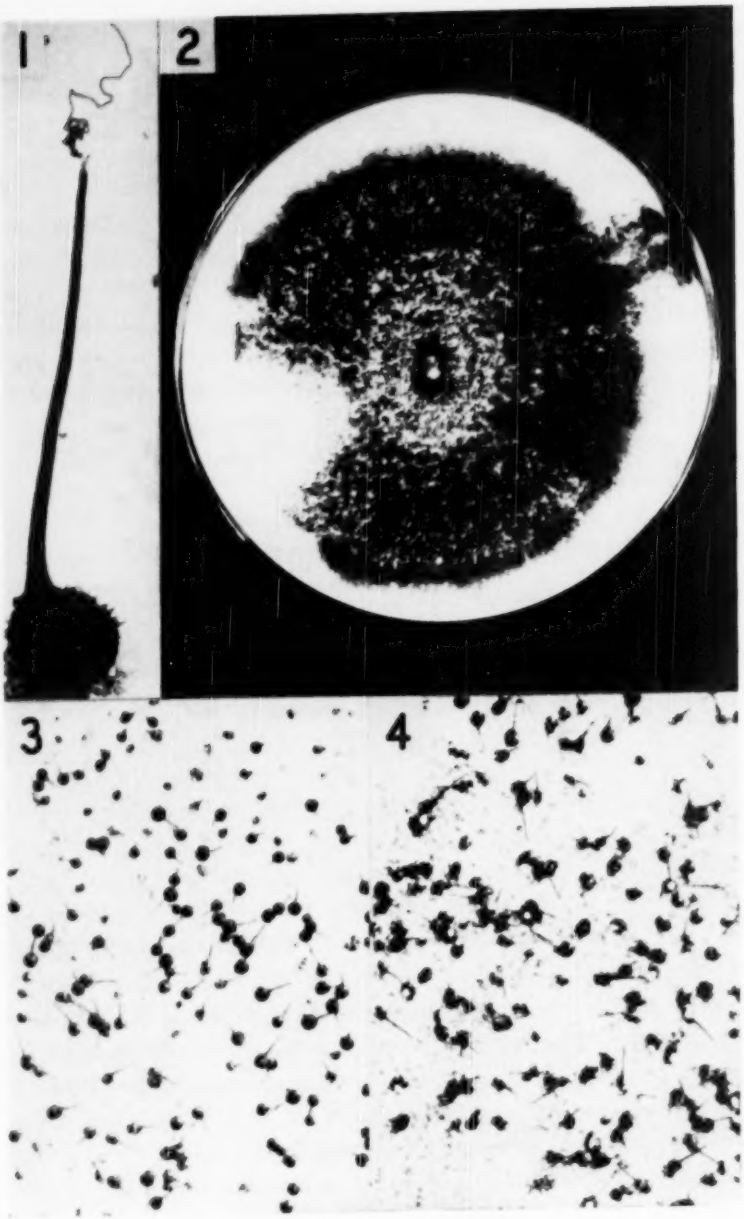
All chemicals were of C.P. or reagent grade unless otherwise specified.

Glassware was cleaned by treatment with dichromate-sulfuric acid solution and rinsed with tap and distilled water.

Further details of method for separate phases of the work will appear in the appropriate ensuing sections.

EXPERIMENTAL RESULTS

Identity of the wood-staining organism: The mold concerned produces the typical long-beaked carbonaceous perithecia (FIG. 1) characteristic of the family Ceratostomataceae. In figure 2 are illustrated the abundant perithecia obtained when this mold was cultured on malt agar. In such cultures the white mycelium becomes darker with age, turning tan and finally dark brown. As is shown in figures 3 and 4, the bulbous base of the perithecium is formed first and, as this matures, the elongate beak grows out therefrom. The beak may be oriented perpendicularly but more often lies at various angles to the substrate, frequently parallel to it. The perithecial base is studded with dark brown to black spines (FIG. 5). Ascus walls are either evanescent or entirely absent, for the ascospores exude single-file through the beak (FIG. 6), pushing aside the fimbriate apical fringe and remaining agglutinated, in either air or an aqueous mounting medium, by virtue of their gelatinous capsules. The spores are ovoid in shape but when unstained have a derby-hat appearance on account of the peculiar capsule. The delimitation of this outer coat is apparent in figure 7, showing spores which have been differentially stained with cotton blue.



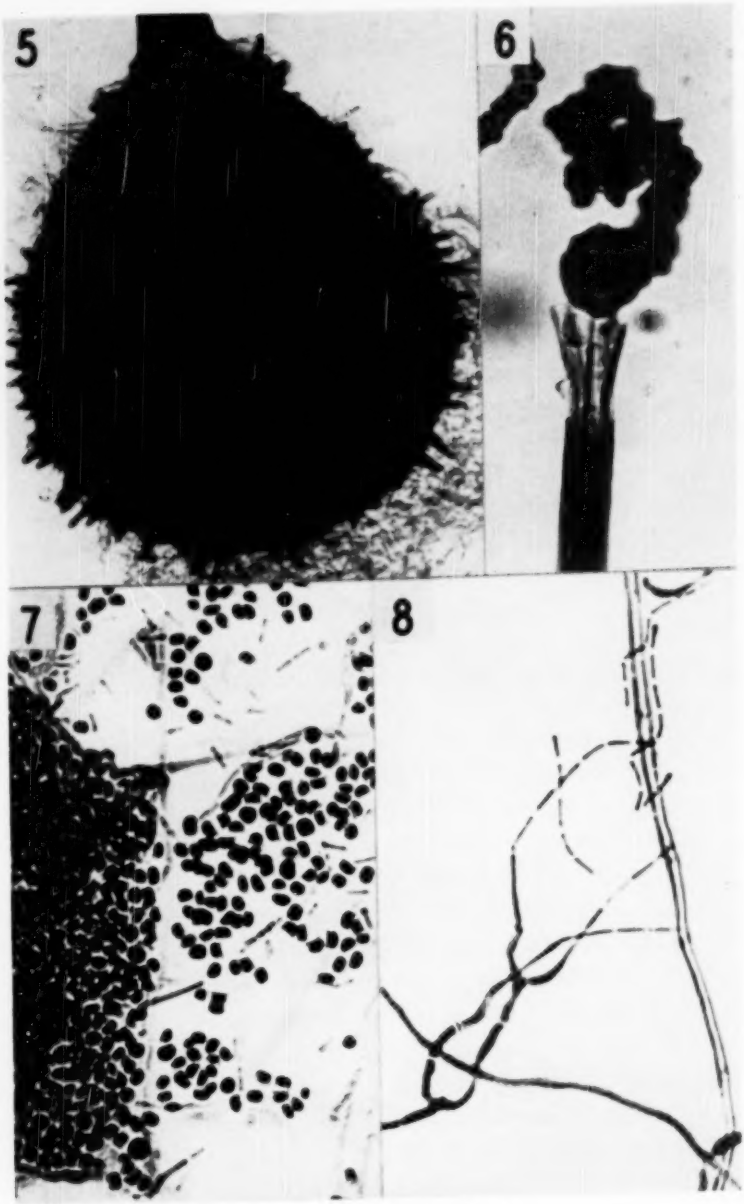
FIGS. 1–4. Blue stain fungi.

The imperfect stage of this mold is distinctive in that conidia are formed endogenously and extruded seriatim from the conidiophore in long moniliform chains (FIG. 8). The appearance of the conidial stage, combined with the characteristics enumerated above, defines this organism as *Endoconidiophora moniliformis* (Hedgc.) Davidson.

For purposes of comparison, a subculture of the organism studied by Davidson and used by him in describing *E. moniliformis* was obtained from Baarn, Holland. This culture was devoid of perithecia, but the two cultures were identical with respect to mycelial growth, type of conidiophore, and size and shape of endoconidia. Although two kinds of endoconidiophores had been described for *E. moniliformis*, only one, the long attenuated variety, appeared in subcultures of the Baarn specimen and in this respect it agreed entirely with the new isolate. When grown on 2.5 per cent Difco malt extract agar, each yielded narrow, hyaline, cylindric conidia which subsequently enlarged, many becoming barrel-shaped and eventually yeast-shaped. Finally, each culture produced a "banana-oil" odor indistinguishable from that of the other.

Basic conditions for growth of E. moniliformis and production of the ester odor: E. moniliformis made good growth, including production of both conidial and perithecial stages, on malt agar, Sabouraud's agar, nutrient agar, carrot-sucrose agar and potato-dextrose agar, when incubated at room temperature (20–25° C.) with no attempt at control of illumination. There was little mycelial growth on corn meal agar, but a few perithecia were produced. The characteristic odor was produced on all of these media.

The following chemically defined medium, essentially that employed by Robbins and Ma (**14**) in their study of the vitamin requirements of certain species of *Ceratostomella*, was, when supplied with 0.4 micromole of thiamin (GBI thiamin hydrochloride) per liter, found to compare very favorably with malt extract media in ability to promote growth, production of perithecia, and formation of the characteristic odor by *E. moniliformis:* dextrose 50 g., KH_2PO_4 1.5 g., $MgSO_4 \cdot 7H_2O$ 0.5 g., asparagin 2.0 g., H_2O q.s. 1000 ml., plus the following trace elements in parts per million: 0.005 B, 0.02 Cu, 0.1 Fe, 0.01 Mn, 0.01 Mo, and 0.09 Zn. This



FIGS. 5–8. Blue stain fungi.

solution, thiamin included, will be referred to hereinafter as the "basal medium."

It was found in preliminary experiments that this mold is not deficient for either biotin or pyridoxine.

The optimum temperature for diameter growth of the present isolate of *E. moniliformis* on both malt agar and the basal medium solidified with agar is approximately 25° C. At this temperature, the average colony diameter on the former medium was 77 mm. and on the latter 85 mm., after 4 days. Amount of odor production appeared to be correlated with diameter growth at each temperature. At 20° C. growth was only slightly less than the optimum and, since a constant-temperature room at 20° was available, all subsequent cultures were incubated at this temperature.

E. moniliformis grows well over a wide range of hydrogen-ion concentrations, producing the ester odor at concentrations between pH 4.22 and pH 6.66 on various synthetic media. All media herein employed were adjusted to pH 4.5–5.0, except as otherwise indicated, this being about the range of pH for different batches of the unadjusted basal medium.

Relation of ester production to growth with respect to time: In order to follow quantitatively the course of ester production and to fix the time of its maximum occurrence in these media, a number of flasks of both malt extract solution (2.5 per cent) and the basal medium, in 100 ml. quantities, were prepared and each inoculated with 0.5 ml. of spore suspension. At frequent intervals, usually of 2–4 days, one flask of each medium was harvested and titrated for total esters (expressed as ethyl acetate; 1 ml. of 0.1 N alkali = 0.0088 g. ethyl acetate). Parallel with this, a time-growth series was run on both media, in which dry weight of mycelium and pH of filtrate were determined for serial samples.

Results are represented by a graph in figure 9, where values for pH and dry weight represent the average for triplicate samples inoculated with 0.1 ml. of spore suspension and grown on 25 ml. of medium in 125 ml. flasks. It is seen that maximum ester production on the basal medium occurs at about 12 days and is closely correlated with maximum dry weight. This suggests that after the original carbon source is exhausted the fungus utilizes the

ester which it has produced. There appears to be no such simple relationship on the more complex malt medium.

Nitrogen requirements of E. moniliformis with respect to growth and ester production: A series of experiments designed to elicit some clue as to the mechanism of formation of the ester was undertaken. Lampitt (**10**) showed that microorganisms (*Saccharomyces cerevisiae*) could deaminate asparagin with the formation of ethyl alcohol. Since this transformation might be involved in the present chemical sequence, it was thought desirable to vary the nitrogen source in the basal medium in order to determine whether asparagin is essential to the manufacture of the ester. If asparagin could be eliminated in favor of a simpler nitrogen

TABLE 1

COMPARATIVE GROWTH, QUALITY OF ESTER ODOR AND QUANTITY OF TITRATABLE ESTERS PRODUCED BY *E. moniliformis* WHEN SUPPLIED WITH VARIOUS NITROGEN SOURCES IN A BASAL MEDIUM CONTAINING 5.0 PER CENT DEXTROSE AS A CARBON SOURCE

Nitrogen compound in g./l.		[a] Comparative growth	[a] Quality of ester odor	[b] Mg. ester per 50 ml. of medium
Control—no added nitrogen		No growth	—	
Inorganic:				
KNO_3	3.00	++	None	
KNO_2 [c]	2.55	Inhibition	—	
NH_4NO_3	1.20	++	Very weak	
$(NH_4)_2SO_4$	1.98	++	Fair	
$(NH_4)_2C_2O_4 + H_2O$	1.86	+++	Good	
$(NH_4)_6Mo_7O_{24} + 4H_2O$	1.85	Inhibition	—	
$NH_2OH \cdot HCl$	2.08	Inhibition	—	
Amides:				
Asparagin	1.98	++++	Good	62.8–18 days
Urea [c]	0.90	++++	Good	89.3–12 days
Amino acids:				
Glutamic acid, Merck	4.41	+++	Good	
Aminoacetic acid, Merck	2.25	++++	Fair	
B-Alanine, Merck	2.67	+	None	
dl-Alanine, Eastman	2.67	++++	Good	
l (-) Leucine, Eastman	3.93	++++	Good	56.3–18 days
l-Cystine, Eastman [d]		+	None	
l-Tyrosine [d]		++++	Fair	
Bactopeptone	2.60	++++	Good	
Malt extract	2.70	++++	Fair	

[a] After 14 days; average of 2–3 flasks.
[b] Each determination on one flask; expressed as ethyl acetate: No. of days as noted.
[c] These compounds sterilized separately by filtration.
[d] Dissolved to limit of solubility.

compound, then glucose could be regarded as the only initial material from which the ester was derived.

Seventeen compounds, containing nitrogen in various valence forms, were substituted individually in equimolar nitrogen quantities for asparagin in the basal medium. In addition, a control set with asparagin and one with only atmospheric nitrogen available were employed. Each solution was prepared in triplicate 100 ml.

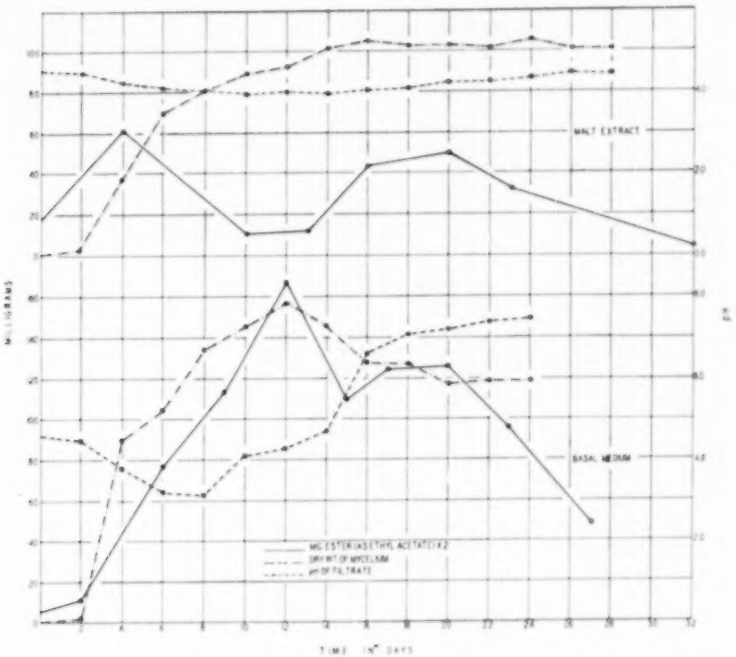


FIG. 9. Growth and ester production by *E. moniliformis* on two media, with corresponding changes in pH of the solutions.

quantities distributed to 250 ml. flasks and 0.5 ml. of spore suspension was used as inoculum. Table 1 shows comparative growth, odor production and, in a few instances where the odor was considered particularly promising, ester titrations for these media.

The inorganic nitrogen compounds employed permitted little or no growth of *E. moniliformis* under the conditions of this experiment, three of them, potassium nitrite, ammonium molybdate, and hydroxylamine hydrochloride, preventing germination when

added in the stated amounts. (In preliminary experiments, conidia and ascospores of this organism germinated readily in distilled water.) Of the organic nitrogen sources tested, urea, peptone, malt extract, and several amino acids permitted mycelial production comparable to that obtained when asparagin was in-

TABLE 2

COMPARATIVE GROWTH, QUALITY OF ESTER ODOR AND QUANTITY OF TITRATABLE ESTERS PRODUCED BY *E. moniliformis* WHEN SUPPLIED WITH VARIOUS CARBON SOURCES IN A BASAL MEDIUM CONTAINING 0.09 PER CENT UREA AS A NITROGEN SOURCE

Carbon compound in g./l.		[a] Comparative growth	[a] Quality of ester odor	[b] Mg. ester per 50 ml. of medium
Control—no added carbon		No growth	—	
Soluble starch	45.0	++++	Good	
Cellobiose	50.0	++++	Fair	16.2–13 days
Dextrose (pH 4.39)	50.0	++++	Excellent	88.6–11 days
Mannose	50.0	++++	Good	74.6–9 days
Galactose	50.0	++++	Doubtful	15.0–13 days
Arabinose	50.0	++++	None	
Xylose	50.0	+++	None	
Glycerol	25.0	+	None	
Calcium lactate	50.0	No growth	—	
Pyruvic acid [c,d] (pH 5.38)	4.0	++	None	
Pyruvic acid [c,d]	1.0	++	None	
Acetaldehyde [c,e] (pH 5.18)	5.0	Inhibition	—	
Acetaldehyde [c,e]	1.0	No growth	—	
Ethyl Alcohol [c]	10.0	++++	Good	72.4–15 days
Ethyl Alcohol [c]	5.0	++++	Fair	
Acetic acid [f]	10.0	Inhibition	—	
Ethyl alcohol [c] + Acetic acid [f] (1:1)	10.0	Inhibition	—	
Ethyl alcohol [c] + Acetic acid [f] (1:1)	5.0	+++	Good	
Ethyl acetate [c]	2.0	++	None	

[a] Estimated after 14 days.
[b] Each determination on one flask; expressed as ethyl acetate; No. of days as noted.
[c] These compounds sterilized separately by filtration.
[d] Added as sodium pyruvate, prepared from Pyruvic Acid, Eastman.
[e] Freshly distilled from Paraldehyde, Eastman.
[f] Added as sodium acetate.

corporated as the nitrogen compound. Urea, furthermore, appeared to be at least the equal of asparagin in permitting production of the ester and had the additional advantage, for use in the further study of the phase-sequence concerned, that it is a relatively simple substance and one not likely to be involved directly in the synthesis.

Carbon requirements of E. moniliformis with respect to growth and ester production: With urea (0.9 g. per l.) as the nitrogen source, various carbon compounds were substituted for glucose in the basal medium in an effort to clarify further the mechanism of ester formation. 100 ml. of media were distributed in triplicate to 250 ml. flasks and inoculated with 0.5 ml. of spore suspension. The results are presented in table 2. Values for growth and for quality of odor represent the average for at least 2 flasks.

Soluble starch, the disaccharide cellobiose, and the hexoses, mannose and galactose, supported good growth of *E. moniliformis*, approximately equal to that on media containing dextrose, but only with starch and mannose, of these compounds, was the ester odor considered good. The 2 pentoses, arabinose and xylose, yielded good to fair mycelial production, but no trace of the ester aroma. Of the 3-carbon compounds employed which are commonly involved in anaerobic respiration and alcoholic fermentation, glycerol and pyruvic acid permitted little growth and no acetate odor, while no growth was observed with lactic acid as the carbon source. Acetaldehyde did not support growth when employed at a concentration of 0.1 per cent, and 0.5 per cent of this compound inhibited germination. Ethyl alcohol, however, served as a good carbon source for production of both mycelial mat and ester, although the yield of both was either impaired or completely prevented by the addition of acetic acid to the alcohol medium. Acetic acid itself, in a concentration of 1.0 per cent, inhibited germination of the spores. Ethyl acetate, 0.2 per cent, when used as a sole carbon source, permitted a small amount of mycelial growth.

Isolation and identification of ethyl acetate: 1) From culture on malt extract: In an attempt to isolate and characterize the substances responsible for the banana oil odor, 102 one-liter flasks, each containing 350 ml. of 2 per cent malt extract solution, were inoculated with 2.0 ml. of spore suspension per flask. The initial pH of this medium was 4.58. Several flasks were harvested each day from the fourth day following inoculation until the twelfth day, when the last batch was processed, each group of cultures being filtered through cheesecloth and the fungus mat and filter cloth stored in a refrigerator. The filtrate

was saturated with sodium chloride and extracted with 4 portions of ether. These were then combined, washed with a portion of 10 per cent Na_2CO_3 followed by several portions of distilled water saturated with NaCl, and finally with 50 per cent $CaCl_2$ solution, then dried over anhydrous $MgSO_4$. The collected mats and filter cloths were steam-distilled and 4 liters of distillate were subjected to ether extraction as above. The dried ether extractives were combined and most of the ether evaporated off through a long fractionating column. The residue was then distilled from a small flask and the following fractions obtained:

1) 34–73° C.: ca. 4.0 ml.
2) 73–82° C.: ca. 0.5 ml; odor like ethyl acetate.
3) 83–107° C.: ca. 0.5 ml.; odor like ethyl acetate.
4) Residue: 0.5 ml.

The first 3 fractions were recombined and fractionated through a semi-micro vigreux column, yielding the following new fractions.

a) 36–60° C.: ca. 2.0 ml.
b) 70–77° C. (Mainly 77°): ca. 0.5 ml.; odor of ethyl acetate.
c) Residue: less than 0.5 ml.

Fraction b) (0.4086 g.) was hydrolyzed by boiling with an excess of N/1 alcoholic NaOH for 1½ hours under reflux. Excess NaOH was titrated with 0.25 N HCl to aqueous phenolphthalein. Saponification equivalent = 96.92. The neutralized solution was distilled and the distillate gave a positive iodoform test at 60° C. (not in the cold).

Thus the odorous substance is probably ethyl acetate.

Residues 4) and c) were combined and subjected to the test for fusel oil as described by Basset (2). The result was negative. Preliminary control runs showed that this test is negative for methyl and ethyl alcohols in concentrations of 1:100, and positive for n-propyl, iso-propyl, n-butyl, sec-butyl, tert-butyl, n-amyl, iso-amyl, and tert-amyl alcohols at the same concentration. N-amyl acetate at a concentration of 1:5000 gave a positive test. The

higher-boiling fractions therefore contained no significant quantity of acetates of the 3-to-5-carbon alcohols.

2) From culture on the basal medium: A similar recovery procedure was followed with 104 flasks, each containing 350 ml. of the basal medium. All flasks were harvested after 12 days and the mats, instead of being steam-distilled, were ground up and extracted directly. After removal of the ether, a total of 3 ml. of distillate was collected in the range 70–78° C. This distillate had a well-defined ethyl acetate odor, but no further purification or characterization of the fraction was attempted.

A more efficient method for recovering the ester was provided by the use of vacuum distillation. Materials for use in vacuum distillation were secured by the following procedure: Loosely wadded cotton balls, 3–4 cm. in diameter, were soaked in the basal medium and excess liquid was expressed by light pressure of the fingers. Seven one-liter flasks were half-filled with these cotton balls and autoclaved. For each flask, 2 malt agar slant cultures were flooded with sterile distilled water, the spore suspensions poured into 50 ml. of basal medium and the mixture distributed aseptically over the flask contents. Incubation was at room temperature (20–25° C.) for 7–10 days. One flask was inoculated per day for 7 successive days and the cultures were harvested serially. As the contents of each flask matured (detected by odor), they were transferred to a vacuum oven connected in series with 2 dry ice-alcohol traps and a high-vacuum pump. Suction was applied continuously for 8 hours and the vacuum maintained overnight. In two instances during this run, several ml. of distillate reached the second trap, and in each case appeared there in 2 layers. The upper layer in each case was separated, the 2 combined, washed with 50 per cent $CaCl_2$ and dried over anhydrous Na_2SO_4. The liquid was then poured off into a side-arm flask and distilled, practically the entire contents (1.5 ml.) coming over at 71–81° C. (fraction 1a).

The remainder of the vacuum distillate from the entire run was extracted with several portions of ether, the ether solution being then washed with 50 per cent $CaCl_2$ and dried over anhydrous

Na_2SO_4. Following evaporation of most of the ether, the residue was distilled and the following fractions collected:

1b) 33–65° C.: ca. 2 ml.
2b) 65–80° C. (mainly 77–80° C.): ca. 2 ml.

Fractions 1a) and 2b) were combined and refractionated through a semi-micro vigreux column, yielding the following new fractions:

1c) 32–74° C.: ca. 1.5 ml.
2c) 74–77.5° C. (mainly 77.1°): ca. 2 ml.; odor of ethyl acetate.

0.3658 g. of fraction 2c) was hydrolyzed by boiling with excess N/1 NaOH for 65 minutes under reflux. Excess NaOH was titrated with 0.1 N HCl to aqueous phenolphthalein. Saponification equivalent = 101.9. The neutralized solution was distilled and the distillate gave a positive iodoform test when warmed to 60° C. Further determinations on fraction 2c): micro boiling-point 77.0° C.; refractive index at 18.9° = 1.3722 (ethyl acetate = 1.37216. Handbook of Physics and Chemistry, 28th Edition).

3) From culture on basal medium modified by replacement of carbon and nitrogen sources: In order to establish the identity of the ester produced in the dextrose-urea and alcohol-urea media, the following procedures were employed. Seven liters of the former medium were prepared and distributed in 350 ml. amounts to 20 one-liter flasks. Initial pH was 4.22. Each flask was inoculated with 2.0 ml. of spore suspension and incubated at 20° C. for 13 days. The entire contents of the flasks were then blended in a Waring Blendor, and the resulting mixture distilled through an 18-inch fractionating column. Two fractions were collected:

1) 70–97° C.: 30 ml. consisting of 2 layers.
2) 97–98.2° C.: 80 ml. consisting of 2 layers.

The upper layers were drawn off and combined (total 6.5 ml.), shaken with an equal volume of 50 per cent $CaCl_2$ solution, separated, and dried over anhydrous $MgSO_4$. The remainder of the collected distillate was then refractionated and 3 fractions obtained:

1a) 69–73° C.: upper layer of 6 ml., water insoluble; lower layer (aqueous), less than 0.5 ml.

2a) 76.5–78.5° C. (mainly 78°): 4 ml. aqueous.
3a) 80.0–99.0° C. (gradual rise): 5 ml. aqueous.

Fraction 1a) was shaken with an equal volume of 50 per cent $CaCl_2$ solution, separated, and dried over anhydrous $MgSO_4$. It was then combined with the upper layers of fractions 1) and 2) and distilled from a small Spitz-type fractionating apparatus, practically the entire volume coming over in the range 70.0–79.8° C. This was collected in four parts:

a) 70.0–74.9° C.: 4 ml.; odor like ethyl acetate.
b) 75.0–76.9° C.: 3 ml.: odor of ethyl acetate.
c) 77.0–77.1° C.: 1 ml.: odor of ethyl acetate.
d) 77.5–79.8° C.: ca. 2 ml.; odor like ethyl acetate.

0.2010 g. of fraction c) was hydrolyzed by boiling with an excess of N/1 alcoholic NaOH for 2 hours under reflux. Excess NaOH was titrated with 0.25 N HCl to aqueous phenolphthalein. Saponification equivalent = 97.6.

A portion of fraction b) was subjected to an elementary analysis by the sodium fusion method (Shriner and Fuson, **15**) and gave negative tests for nitrogen, sulfur and halogens. A second portion was treated with 3,5-dinitrobenzoyl chloride and sulfuric acid and the resulting 3,5-dinitrobenzoate recrystallized from ethanol. The crystals melted at 92.9° C. (ethyl ester of 3,5-dinitrobenzoic acid = 93.0° C.). A third portion was refluxed with hydrazine hydrate and the hydrazide recrystallized from a water-ethanol mixture. The purified substance melted at 66.9° C. (hydrazide of acetic acid = 67° C.). The production of ethyl acetate is thus confirmed.

Fractions 2a) and 3a) were shown to contain ethyl alcohol by the following procedure. Two ml. of the liquid were treated with 3,5-dinitrobenzoyl chloride and the resulting precipitate recrystallized from a water-ethanol mixture. Melting point = 93.0° C.

Ten one-liter flasks, each containing 350 ml. of the alcohol-urea solution, were inoculated with 2.0 ml. of spore suspension each, incubated at 20° C. and harvested after 15 days. Treatment was the same as that described for the dextrose-urea medium, resulting in the recovery of 6 ml. of distillate in the range 70.0–80.0° C. S.E. on 75.0–77.1° fraction = 95.0. Elementary analysis negative

for nitrogen, sulfur and halogens. Odor of ethyl acetate. M.P. of 3,5-dinitrobenzoate = 92.8° C. M.P. of hydrazide = 66.9° C. Ethyl acetate is therefore produced from ethyl alcohol as a carbon source, in the presence of urea as a nitrogen source.

DISCUSSION

On media in which ethyl acetate is formed, the amount produced apparently parallels the growth of the mycelium. This, together with the fact that *E. moniliformis* can utilize ethyl acetate as a sole carbon source, indicates that the ester is utilized only after more readily available nutrients are exhausted. Acetic acid is apparently toxic to the organism, even preventing germination when employed as the carbon source in a synthetic medium. Its esterification by ethyl alcohol manifestly serves as a mechanism whereby the toxicity of the acid is removed. It would appear that the oxidation of ethyl alcohol to acetic acid and subsequent enzymatic coupling of these two compounds comprise the final steps in the phase-sequence of ethyl acetate production. Since acetaldehyde in relatively low concentrations supports no growth and even inhibits germination, its possible role here as an intermediary substance in the manufacture of ethyl acetate must be considered doubtful. Lactic acid, pyruvic acid, and glycerol, substances which are involved in ordinary anaerobic fermentation, also permit little or no growth of *E. moniliformis*. Xylose and arabinose, although serving well as carbon sources for mycelial production, yielded no detectable odor of ethyl acetate, whence it may be assumed that little or none was produced. It is probable, therefore, that no significant amount of alcohol was generated from these sugars, although *Fusarium lini* Bolley was shown by White and Willaman (**16, 17**) to produce ethyl alcohol from pentoses as well as from hexoses and apparently via similar pathways.

SUMMARY

1. A mold causing a blue stain of pine wood, identified as *Endoconidiophora moniliformis* (Hedgc.) Davidson, grows well in a solution of mineral salts, dextrose, and asparagin, when supplied with thiamin. It has a wide range of pH tolerance on this and

other media, and an optimum temperature for mycelial growth of approximately 25° C.

2. Considerable quantities of ethyl acetate as well as some ethyl alcohol are produced in the above solution, and the ester is formed also in malt extract solution, but no higher acetates were detected in cultures on either medium.

3. The production of esters on the dextrose-asparagin medium is correlated closely with gain in dry weight by the fungus, a maximum for both being attained in about 12 days at a temperature of 20° C., following which the esters are gradually decomposed.

4. *E. moniliformis* produces good growth, accompanied by varying amounts of ester, on the above medium when asparagin is replaced by any of several organic nitrogen compounds, notably urea, but in general neither growth nor ester production is pronounced when inorganic nitrogen sources are used. The dextrose-urea medium yields relatively large quantities of ethyl acetate.

5. When urea is employed as the nitrogen source and other carbon compounds substituted for dextrose in the medium, it is found that soluble starch, mannose, cellobiose, and galactose permit good growth and good to poor ester production, and 2 pentoses good growth but no ester odor. Various 3-carbon intermediates in the usual chain of alcoholic fermentation support little or no growth and yield no detectable ester odor. Acetic acid in relatively low concentrations inhibits germination and growth of the organism, but ethyl acetate is utilized for the production of mycelium.

Ethyl alcohol serves as an excellent carbon source for *E. moniliformis* with respect to both growth and production of ethyl acetate.

6. The significance of these findings for the elucidation of the phase-sequence of ethyl acetate synthesis by *E. moniliformis* is discussed.

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EXPLANATION OF FIGURES

FIG. 1. Perithecium extruding ascospores. Stained with cotton blue (×92).

FIG. 2. Malt agar plate culture: numerous perithecia; white mycelial growth on periphery (×4/5).

FIG. 3. Malt agar plate culture: stages in development of perithecia (×8).

FIG. 4. Malt agar plate culture: mature perithecia with apical clumps of ascospores (×8).

FIG. 5. Base of perithecium: numerous spines (×256).

FIG. 6. Mouth of perithecium: ascospores being extruded. Stained with cotton blue (×404).

FIG. 7. Mass of ascospores, showing gelatinous capsules. Stained with cotton blue (×556).

FIG. 8. Conidiophores and endoconidia. Stained with cotton blue (×556).

NOTES ON TWO LITTLE KNOWN BIRD'S NEST FUNGI FROM SOUTHERN UNITED STATES

HAROLD J. BRODIE

(WITH 2 FIGURES)

As was pointed out by Lloyd, there are but four common and widely distributed species of the Nidulariaceae in the United States. Lloyd (3) reported that of one hundred and seventy-six specimens he had received from correspondents, one hundred and sixty-five belonged to one or other of the following: *Crucibulum vulgare* Tul., *Cyathus stercoreus* (Schw.) De Toni, *C. striatus* Willd., *C. vernicosus* DC.

The purpose of this note is to draw attention to two species from southern United States, one of which appears to be scantily represented in the herbaria of the southern states, the other to be known within the United States from only a single collection. It is believed that these species might be more frequently collected if their characteristics were better known and both are therefore illustrated and described in this paper.

CYATHUS POEPPIGII Tul.

In Miss White's monograph (4) this *Cyathus* was reported from the Danish West Indies. It was originally described from Cuba. Lloyd (3) lists collections from Mauritius, German East Africa, Australia and Samoa. Coker and Couch (1) refer to the first collection of this species in the United States from Gainesville, Florida, 1924. Some months ago, the writer examined some specimens of *C. Poeppigii* collected by Dr. Erdman West at Gainesville, Jan. 16, 1941. Dr. L. R. Hesler, who kindly loaned the specimens from the Herbarium of the University of Tennessee, was of the opinion that this species had also been found in Tennessee.

West's collection consists of some two dozen fruit bodies that had evidently developed in clusters. The cups are *dark chocolate*

brown, about 8 mm. high and 5–6 mm. wide at the mouth. Superficially they might be mistaken for small depauperate forms of *C. striatus*. *C. Poeppigii* is distinguished macroscopically by the striations or grooves on the upper one-third of the *outside* of the cups (FIG. 1). However, because the young fruit bodies are hirsute, the external striation is apt to be overlooked. Moreover, the writer has examined more than one collection of *C. striatus* in which there is slight striation on the outside of the cup. One such collection from Indiana was actually labelled "*C. Poeppigii*" by the collector. There is *no tunica* covering the mature peridioles of *C. Poeppigii* and these therefore appear very dark brown or black and shiny (FIG. 1). The basidiospores are *very large*, measuring 22–25 × 30–32 μ in the West collection. The spore size, of course, distinguishes this species very readily from *C. striatus*. A comparison of the characteristics of *C. Poeppigii* and *C. striatus* is given in table 1.

TABLE 1

COMPARISON OF *Cyathus Poeppigii* AND *C. striatus*

	Cyathus Poeppigii	*Cyathus striatus*
Size	8–10 mm. high; 5–6 mm. wide at mouth.	8–12 mm. high; 6–8 mm. wide at mouth.
Shape	Broad—conic.	Usually more elongate, conic.
Color	Chocolate brown.	Light to medium brown although darker forms occur.
Outer Surface	Shaggy or hirsute; hairs around mouth inconspicuous.	Very hirsute; hairs around mouth usually very conspicuous.
Striation	Marked *external* striation as well as internal.	Striae *internal*; if external, very inconspicuous.
Peridioles	Lacking tunica; dark brown to black; 1.5–2 mm. in diameter.	Usually with evident light-colored tunica.
Basidiospores	Large, variable; mostly 22–25 × 30–35 μ.	Small, variable; mostly 8–12 × 18–20 μ.

CYATHUS PALLIDUS Berk. and Curt.

As far as the writer has been able to ascertain, this species has been collected in the United States only once. It was found by

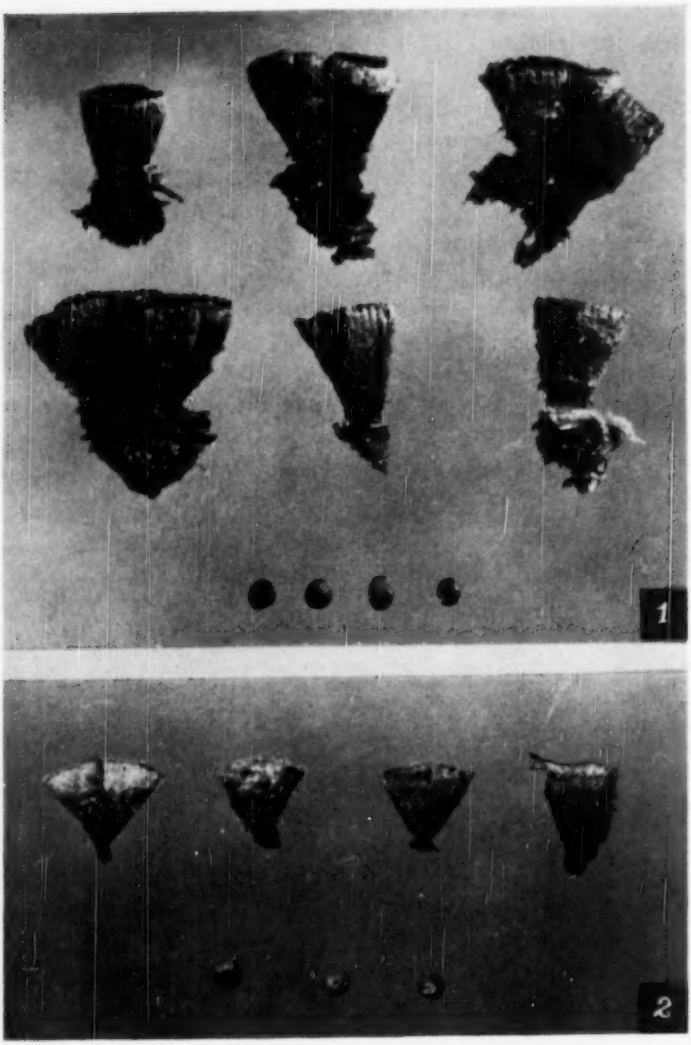

FIG. 1. *Cyathus Poeppigii* fruit bodies and peridioles. The grooves or striae on the outside of the peridium and the dark shiny peridioles devoid of tunica are characteristic of this species. Specimens from a collection made by Dr. Erdman West, Gainesville, Fla., Jan. 16, 1941. ×2.

FIG. 2. *Cyathus pallidus* fruit bodies and peridioles. The two specimens on the left show the coarse hairs characteristic of this species. Specimens collected by Dr. William Diehl, Savannah, Ga., Jan. 6, 1933. ×1⅓.

Dr. William Diehl (Jan. 6, 1933) at Savannah, Georgia, and reported by him in Mycologia, 1941 (2). The specimens were growing on decaying boxes that had formerly contained cuttings of camellia plants. The species was originally described from Cuba and was reported by Lloyd (3) from Jamaica and Antigua and by Miss White (4) from Cuba.

Dr. Diehl kindly allowed the writer to examine his collection of *C. pallidus* and the following notes are based upon this examination. The four specimens of this fungus (FIG. 2) are *pale buff* or

TABLE 2

COMPARISON OF *Cyathus pallidus* AND *C. stercoreus*

	Cyathus pallidus	*Cyathus stercoreus*
Size	7–9 mm. high; 8–9 mm. wide at mouth.	Variable, from 5–10 mm. high; strains as small as *C. pallidus* occur.
Shape	Broad—conic.	Variable but usually with sides less divergent than *C. pallidus*.
Color	Pale gray-brown.	Brown or gray.
Outer Surface	Hispid with numerous rigid hairs.	Very shaggy with matted hairs.
Peridioles	With delicate tunica; 2 mm. in diameter.	Devoid of tunica; dark brown or black; 2 mm. in diameter.
Basidiospores	Small; subglobose to elliptical; 5–7 × 8–10 μ.	Large; subglobose; 30–35 μ.

fawn-colored, 7–9 mm. high and 8–9 mm. wide at the mouth. In shape they are widely conical. The peridium is *thin* and membranous and is covered externally with hairs. In addition there are *coarser rigid hairs* that are quite conspicuous. In Dr. Diehl's specimens, the cups are very faintly ridged on the inner surface. Lloyd (3) commented: "The type specimens appear *very slightly striate*, but the plant belongs in the section Olla, not in Eucyathus as found in Saccardo."[1] The peridioles are 2 mm. in diameter and have a *thin tunica*. Basidiospores are *small*, mostly 7 × 8.5 μ in the Diehl specimens, thick-walled and colorless.

[1] The italics are Lloyd's.

Because this species is not unlike several collections from Florida of a small gray form of *C. stercoreus* (except for the large spores of the latter) which the writer has seen, a comparison of the two species is given herewith (table 2).

The superficial resemblance of *Cyathus Poeppigii* to small dark forms of *C. striatus*, and of *C. pallidus* to small pale forms of *C. stercoreus* may have resulted in the two rare species being overlooked by collectors. The appearance of these fungi in Georgia and Florida suggests their introduction from the adjacent tropics. Dr. Diehl (2) found *C. pallidus* growing underneath camellia cuttings in a commercial nursery and Dr. West found *C. Poeppigii* at Gainesville, Fla.

DEPARTMENT OF BOTANY,
INDIANA UNIVERSITY,
BLOOMINGTON, INDIANA

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2. **Diehl, W. W.** 1941. The taxonomy of Zenker's *Leptostroma Camelliae*. Mycologia **33**: 215–219.
3. **Lloyd, C. G.** 1906. The Nidulariaceae. Cincinnati. 32 pp.
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NOTES AND BRIEF ARTICLES

ANNUAL FORAY, 1948

Inasmuch as the A.A.A.S. had announced plans for an autumn meeting for 1948, it was decided to hold the annual foray of the Society in the early summer. Such a course had two advantages, namely, there would not be two meetings within a short space of time, and an opportunity would be afforded to collect representatives of the late spring and early summer flora.

Through the kindness of Dr. A. H. Stockard, the director of the University of Michigan Biological Station, the Society was enabled to utilize the facilities of the Station from June 15 to 17. The Station is located in Cheboygan County in the northern part of the southern peninsula of Michigan.

On Tuesday morning, June 15, collections were made in the Carp Creek gorge, an area with a primarily coniferous cover. Identification of material and further collecting at nearby sites occupied the afternoon. In the evening, before a most welcome open fire in the recreation hall, members were treated to an extraordinarily fine display of Kodachrome pictures of fungi by E. B. Mains, A. H. Smith and B. B. Kanouse, of the University Herbarium. Wednesday, June 16, was devoted to an all day trip to the scenic Harbor-Springs-Cross Village area bordering Lake Michigan. Collections were made in the hardwoods bordering the lake. After a picnic lunch, collections were made at other points along the return route to the Biological Station. That evening, Dr. E. B. Mains explained and illustrated a series of stereokodachrome pictures of fleshy fungi which is in preparation. Thursday, June 17, was devoted to collecting in two nearby bogs. In the morning the group went to Mud Lake Bog where a number of interesting finds were made. In the afternoon Reese's Bog, bordering Burt Lake, was the collecting area.

Although an early summer drought did not make collecting as fruitful as it might have been, a goodly number of interesting speci-

Annual foray, 1948, Douglas Lake, Michigan.

mens were brought back to the Station. A list of these together with their collectors is appended.

I would be remiss, indeed, if special mention were not made at this time of the yoeman service performed by A. H. Smith in making the foray a success. His familiarity with the terrain was of tremendous value. Nor were Smith's contributions to the foray confined entirely to fleshy fungi. It was with pardonable pride that the Vice-President noted he was evidently educating certain well-known rust and agaric specialists in the methods of finding aquatic fungi. At least, so it seemed, for every evening at dusk these gentlemen would adjourn to some nearby stream with boots and straw collecting baskets. Since the substrata obtained never seemed to possess saprolegniaceous fungi on them, they were promptly utilized for other worthy but distinctly non-mycological purposes.

The following is a list of the fungi collected. The collector's names are abbreviated and in parentheses, *i.e.*, St. = Stevenson, S. = Sinden, Ri. = Reischer, etc.

LIST OF SPECIES

Achlya flagellata: 4 collections from Maple River, June 15, 1948. 2 from Lake Shore cut off, June 16, 1948. 1 from Mud Lake Bog, June 17, 1948 (Ri.)

Achlya Klebsiana (probably) : 1 collection from Maple River, June 15, 1948 (Ri.)

Achlya papillosa: 2 collections from Mud Lake Bog, June 17, 1948 (Ri.)

Achlya polyandra (probably) : 1 collection from Maple River, June 15, 1948 (Ri.)

Ascochyta cassandrae Pk., on *Chamaedaphne calyculata* (L.) Moench. (S.)

Bifusella Faullii Darker, on *Abies balsamea* (L.) Mill. (S.)

Chrysomyxa cassandrae (P. & C.) Tranz., on *Chamaedaphne calyculata*, Mud Lake Bog, June 17, 1948, Rogerson & JAS. (St.)

Chrysomyxa cassandrae (P. & C.) Tranz., on *Chamaedaphne calyculata* (L.) Moench. (S.)

Chrysomyxa chiogenis Diet., on *Chiogenes hispidula* (L.) T. & G. (S.)

Chrysomyxa ledi (A. & S.) de Bary, on *Ledum groenlandicum* Oeder (S.)

Chrysomyxa ledi (Alb. & Schw.) de By, on *Ledum groenlandicum*, Mud Lake Bog, June 16, 1948 (St.)

Daedalea unicolor Bull. ex Fr., on *Betula* (dead wood), below Cross Village (St.)

Erysiphe graminis D. C., on *Agropyron repens*, Grounds, Biol. Sta., June 15, 1948 (St.)

Fomes pini (Fr.) Karst., on *Picea glauca*, Mud Lake Bog, June 17, 1948, det. J. L. Lowe (St.)

Fomes pinicola (Fr.) Cke., on *Tsuga canadensis*, Gorge, Biol. Sta. U. Mich., June 15, 1948 (St.)

Fomes scutellatus (Schw.) Cke., on *Alnus* (dead twigs), Mud Lake Bog, June 17, 1948 (St.)

Fomes subroseus (Weir) Overh., on *Abies balsamea*, Mud Lake Bog, June 17, 1948 (St.)

Herpobasidium filicinum (Rostr.) Lind., on *Dryopteris disjuncta* (Rupr.) Morton (S.)

Hyalopsora aspidiotus (Pk.) Magn., on *Abies balsamea*, above Harbor Springs, June 16, 1948, Kern et al. on *Phegopteris dryopteris* above Harbor Springs, June 16, 1948, Kern et al. (St.)

Hyalopsora aspidiotus (Pk.) Magn., on *Abies balsamea* (L.) Mill. and on *Dryopteris disjuncta* (Rupr.) Morton (S.)

Hymenochaete tabacina (Sow. ex Lév.), on *Alnus* (dead branches), Mud Lake Bog, June 17, 1948 (St.)

Hypodermella punctata Darker, on needles of *Abies balsamea*, below Cross Village, June 16, 1948; on needles of *Abies balsamea*, Mud Lake Bog, June 17, 1948 (St.)

Irpex cinnamomeus Fr., on *Quercus* (dead branches)—above Harbor Springs, June 16, 1948 (St.)

Isoachlya unispora: 1 collection from Carp Creek, the Gorge, June 15, 1948. 1 from Maple River, June 15, 1948 (Ri.)

Lachnum virgineum (Batsch. ex Fr.) Karst., on dead twigs, Mud Lake Bog, June 17, 1948 (St.)

Leptolegnia caudata: 1 culture from Mud Lake Bog, June 17, 1948 (Ri.)

Leptothyrium periclymeni (Desm.) Sacc. var. *americanum* E. & E., on *Lonicera canadensis* Marsh (S.)

Mycosphaerella chimaphilina (Pk.) House, on *Chimaphila umbellata*, below Cross Village, June 16, 1948 (St.)

Mycosphaerella chimaphilina (Sacc.) House, on *Chimaphila umbellata* (L.) Bart. var. *cisatlantica* Blake (S.)

Peltigera canina (L.) Willd., Reese's Bog, June 17, 1948 (St.)

Peniophora cinerea Fr., on *Betula* (dead twigs), Gorge, Biol. Sta., June 15, 1948 (St.)

Phyllachora Witrockii (Erikss.) Sacc., on *Linnaea borealis* L. var. *americana* (Forbes) Rehder (S.)

Polyporus abietinus Dicks. ex Fr., *Tsuga canadensis*, Gorge, Biol. Sta., June 15, 1948; *Betula* sp., Gorge, Biol. Sta., June 15, 1948 (St.)

Polyporus biformis Kl., on dead wood, below Cross Village, June 16, 1948 (St.)

Puccinia amphigena Diet., on *Calamovilfa longifolia*, above Harbor Springs, Emmet Co., June 16, 1948 (St.)

Puccinia caricis-grossulariata Arth., on *Grossularia*, Maple River, June 15, 1948 (St.)

Puccinia coronata Cda., on *Rhamnus alnifolia*, Reese's Bog, June 17, 1948 (St.)

Puccinia coronata Cda., on *Rhamnus cathartica* L. (S.)

Puccinia extensicola solidaginis (Schw.) Arth., on *Solidago altissimum*, Reese's Bog, June 17, 1948; on *Solidago* sp., Reese's Bog, June 17, 1948 (St.)

Puccinia pringsheimiana Kleb., on *Ribes* sp. (S.)

Puccinia violae (Schum.) D. C. (?), on *Viola* sp., Gorge, Biol. Sta., June 15, 1948 (St.)

Pucciniastrum Pyrolae (Pers.) Schroet., on *Pyrola secunda* L. (S.)

Ramularia arvensis Sacc., on *Potentilla norvegica* L. var. *hirsuta* (Michx.) Lehm. (S.)

Ramularia Magnusiana (Sacc.) Lindau, on *Trientalis borealis* Raf. (S.)

Rhytisma andromedae (Pers.) Fr., on *Andromeda glaucophylla* Lk. (S.)

Saprolegnia ferax: 2 collections from Carp Creek, the Gorge, June 15, 1948 (Ri.)

Saprolegnia litoralis: 2 collections from Carp Creek, the Gorge, June 15, 1948 (Ri.)

Septoria coptidis Berk. & Curt., on *Coptis trifolia*, Mud Lake Bog, June 17, 1948 (St.)

Septoria osmorrhizae Pk., on *Osmorrhiza* sp., Maple River, June 15, 1948, J. C. Gilman (St.)

Septoria trillii Pk., on *Trillium*, above Harbor Springs, June 16, 1948 (St.)

Septoria trillii Pk., on *Trillium grandiflorum* Michx. (S.)

Steccherinum ochraceum (Fr.) S. F. Gray, on *Alnus* (dead bark), Mud Lake Bog, June 17, 1948 (St.)

Stereum fasciatum Schw., on *Betula* (fallen log), below Cross Village, June 16, 1948 (St.)

Stereum sanguinolentum (Alb. & Schw.), on *Abies balsamea*, below Cross Village, June 16, 1948 (St.)

Taphrina ?bacteriosperma Johans., on *Betula papyrifera* Marsh var. *cordifolia* (Regel) Fern. (S.)

Taphrina caerulescens (Mont. & Desm.) Tul., on *Quercus ?borealis* Michx. F. (S.)
Taphrina caerulescens (Mont. & Desm.) Tul., on *Quercus maxima*, Biol. Sta., June 15, 1948 (St.)
Taphrina dearnessii Jenkins, on *Acer rubrum* L. (S.)
Taphrina dearnessii Jenkins, on *Acer rubrum*, Reese's Bog, June 17, 1948, on *Acer rubrum*, also seen at Biol. Sta., June 15, 1948 (St.)
Taphrina flava Farl., on *Betula papyrifera*, Reese's Bog, June 17, 1948 (St.)
Taphrina virginica Seym. Sadeb., on *Ostrya virginiana* (Mill.) Willd. (S.)
Thraustotheca clavata: 1 culture from Mud Lake Bog, June 17, 1948 (Ri.)
Uromyces acuminatus Arth., on *Smilacina stellata*, above Harbor Springs, June 16, 1948 (St.)
Uromyces acuminatus Arth., on *Maianthemum canadense*, above Harbor Springs (Emmet C.), June 16, 1948 (St.)
Uromyces caladii (Schw.) Farl., on *Arisaema triphyllum*, Maple River, June 15, 1948 (St.)
Uromyces caladii (Schw.) Farl., on *Arisaema atrorubens* (Ait.) Blume. (S.)
Venturia dickiei (Berk. & Br.) Ces. & de N., on *Linnaea borealis* L. var. *americana* (Forbes) Rehder (S.)
Venturia dickiei (Berk. & Br.) Ces. & de Not., on *Linnaea borealis*, Reese's Bog, June 17, 1948 (St.)
Venturia pulchella C. & P., on *Chamaedaphne calyculata* (L.) Moench. (S.)

—F. K. SPARROW.

SHOULD DR. D. P. ROGERS' PROPOSALS FOR THE REGULATION FOR FIXING THE GENERIC TYPES BE ACCEPTED AS PUBLISHED IN MYCOLOGIA **41**: 219, 1949?

The proposals made by Dr. D. P. Rogers regarding the fixation of the generic types are extremely important for further taxonomic work in the fungi as well as in other special fields of botany. It is the writer's impression that they should be discussed widely before they are submitted to the International Congress at Stockholm. As a member of the "Special Committee on Fungi for the International Congress at Stockholm," the writer is expected to either approve or disapprove of proposals submitted, but in the case of Dr. Rogers' proposals, a mere yes or no would not be sufficient. On one hand, this proposal has the merit to give the taxonomist a "clear, simple, and honest" (as Rogers says himself) tool for the selection of types; on the other hand it is too rigid and explicit to satisfy those who insist on keeping all additional rules in line with the basic idea as expressed in Art. 3 (1) "to aim at fixity of

names" and Art. 18, Recommendation VI, "choose a species that will fix the generic name as it is now commonly applied." For example, following the proposed rules as edited by Rogers one would have to exclude the generic name *Cantharellus* and either abandon (as a synonym) or change the sense of the genus *Psathyrella*. Rogers Art. 5(b) would, if interpreted literally, imply that only species with white spores are typical for *Cantharellus* since this is stated in the original description. The typical *Cantharelli* in the sense of all authors have colored (yellow or rosy) spores while those species originally admitted in *Cantharellus* that have white spores, are now excluded from the genus. However, being forced by a rigid rule, one would have to exclude the species generally accepted (Earle, Murrill, Clements & Shear, Singer & Smith, Doty) as generic type, *Cantharellus cibarius*, and admit one of the white-spored species instead which would upset the taxonomy of the whole complex. In the case of *Psathyrella*, one would have to follow the even more dangerous rule indicated in Art. 6 ("Among species equally eligible, the preference shall be given to the first known to have been designated as the type"); this would be *Psathyrella disseminata*, now separated from *Psathyrella* in the sense of all modern authors and either incorporated into *Coprinus*, or type species of a small genus, *Pseudocoprinus*. Consequently, *Psathyrella* would become a synonym of *Coprinus* or it would replace *Pseudocoprinus*, and the species of *Psathyrella*, now more numerous than ever, would have to be transferred to *Hypholoma*, or *Psilocybe*, both used in another sense by some mycologists, and in the sense of *Psathyrella* by none. These examples might be supplemented by others of the same group of fungi as well as by examples from other cryptogams and phanerogams. One can by no means imply that these or similar generic names belong to what Rogers calls "a beloved, but illegitimate nomenclature." In the contrary, the acceptance of the rules proposed by Rogers would tend to "cause error and ambiguity." Rogers' proposals would have been excellent if they had been published 30 years ago. Now, they will help solving some of the problems accumulated because of the absence of such rules, but at the same time, they will destroy that partial structure of sound, acceptable generic names created

by the common sense of many authors and tradition of up to two centuries. Aside from that, one will have to consider the difficulty of establishing definitely the first designation of a type species, and the changes in what is considered disagreement with an original diagnosis.

The writer proposes to use Rogers' proposal (or a similar one) as a basis but restricting the rules to articles 1–3, or possibly 1–5(a), referring Art. 4–6, or at least 5b, and 6 to the recommendations, since such a set of rules and recommendations does not and should not solve every single problem of typification. However, the writer cannot see why the remaining difficulties cannot be overcome by lists of "Species Lectotypicae" such as given in the supplement written by Hitchcock & Green for the Linnean generic names of the Phanerogamae. This has the additional advantage to promote the preparation of lists by specialists in various groups thus avoiding the rigid application of rules which would upset the nomenclature in certain cases. After such lists have been submitted, they should be discussed, and judged by the sincerity with which the author has applied the existing rules and recommendations, and by the respect he has shown for the names based on established custom, as long as they are legitimate.

Whether Appendix I in the end takes the form of Rogers' proposals, or as here proposed, that of a short and simple set of rules and recommendations plus a supplement of lists of "Species Lectotypicae"—let us not forget that action one way or the other is preferable to rejection of or inaction on both proposals.

—ROLF SINGER.

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THE MYXOMYCETES [1]

The embodiment of Dr. G. W. Martin's long-continued studies in the taxonomy of the Myxomycetes has recently appeared as vol-

[1] MARTIN, G. W. Fungi. Myxomycetes. Ceratiomyxales, Liceales, Trichiales, Stemonitales, Physarales. North American Flora **1** (1): 1–151. Bibliography, by H. W. Rickett, pp. 153–178. Index, pp. 179–190. 1949—New York Botanical Garden, $7.25.

ume 1, part 1, of North American Flora. Students of these fungi will note a considerable change from the arrangement of the component groups appearing in Macbride, Macbride & Martin, and Hagelstein, the predecessors of the present work. In addition, a number of small genera and of poorly-marked species have been combined with related groups with the purpose of better expressing the existing relationships.

Although parts of the North American Flora dealing with fungi have been appearing since 1906, it has been left to Dr. Martin to define (on p. 1) the Division *Fungi*.—DONALD P. ROGERS.

Notice to Contributors to Mycologia

Professor G. W. Martin of the Department of Botany of the University of Iowa has been elected to the position of Editor-in-Chief of Mycologia for the five year term beginning with the volume for 1951. After September 1, 1950 all manuscripts should be submitted to him at the above address. Professor Martin expects to be in Europe for some months previous to the above date, so the present incumbent will function until that time. This is important to those who will be submitting papers for the last two issues of 1950 since if such papers are sent to Professor Martin during the summer months they might accidentally be neglected until work is started on the 1951 issue.—ALEXANDER H. SMITH.

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